(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
15 January 2004 (15.01.2004)

PCT

(10) International Publication Number
WO 2004/005522 A2

(51) International Patent Classification[7]: C12N 15/80

(21) International Application Number: PCT/EP2003/007028

(22) International Filing Date: 2 July 2003 (02.07.2003)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
02015067.8 5 July 2002 (05.07.2002) EP
03008444.6 11 April 2003 (11.04.2003) EP

(71) Applicant *(for all designated States except US)*: **BASF AKTIENGESELLSCHAFT** [DE/DE]; ., 67056 Ludwigshafen (DE).

(72) Inventors; and

(75) Inventors/Applicants *(for US only)*: **LACOUR, Thierry** [FR/DE]; Lutherstr. 4, 76297 Stutensee (DE). **RETHER, Jan** [DE/DE]; St.-Marien-Platz 23, 67655 Kaiserslautern (DE). **FREUND, Annette** [DE/DE]; Römerweg 17c, 67117 Limburgerhof (DE). **SCHÄFER, Wilhelm** [DE/DE]; Vörloh 18, 22589 Hamburg (DE). **MAIER, Frank** [DE/DE]; Gehrden 17, 21635 Jork (DE). **MALZ, Sascha** [DE/DE]; Bargfredestr. 10d, 22587 Hamburg (DE).

(74) Common Representative: **BASF AKTIENGESELLSCHAFT**; ., 67056 LUDWIGSHAFEN (DE).

(81) Designated States *(national)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
— *without international search report and to be republished upon receipt of that report*

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

53691
030504

WO 2004/005522 A2

(54) Title: PLASMID VECTORS FOR TRANSFORMATION OF FILAMENTOUS FUNGI

(57) Abstract: The invention relates to novel plasmid vectors for transformation of filamentous fungi and to a method of modifiying the genome of filamentous fungi based on these vectors. The invention furthermore relates to the modification of a specific gene via the process of homologous recombination, to recombinant expression of foreign genes in filamentous fungi and to new selection markers for detecting succesfull transfer of the target gene in filamentous fungi.

ERU

Plasmid vectors for transformation of filamentous fungi

Description

The invention relates to novel plasmid vectors for transformation of filamentous fungi and to a method of modifiying the genome of filamentous fungi based on these vectors. The invention furthermore relates to the modification of a specific gene via the process of homologous recombination, to recombinant expression of foreign genes in filamentous fungi and to new selection markers for detecting the successful transfer of the target gene in filamentous fungi.

One method currently used for transformation of filamentous fungi is random mutagenesis based on transposons insertion, a method also known for plant transformation (WO 01/38509). This method allows the genomes of several species such as Magnaporthe grisea to be studied (for examples WO 00/55346; WO 00/56902). However, this strategy requires a big effort in terms of bioinformatic tools and molecular biology to localise precisely the insertion in the genome.

Alternatively, known transformation methods are based on targeted integration. Targeted transformation of fungi can be carried out either by offering a knock-out cassette with a marker-gene flanked by two homologous sequences (Aronson et al, 1994, Mol. Gen. Genet. 242: 490-494; Royer et al, 1999, Fungal Genetics and Biology 28: 68-78; Schaefer, 2001, Current Opinion in Plant Biology 4: 143-150) or by quoting a plasmid with the marker gene in the neighbourship of a homologous sequence (Shortle et al.,1982, Science 217: 371-373; Bird and Bradshaw, 1997, Mol Gen Genet. 255: 219-225; Feng et al., 2001, Infection and Immunity 69 (3): 1781-1794; Schaefer, 2001, Current Opinion in Plant Biology 4: 143-150). Both procedures are in principle attractive methods to study the gene function, but they have the disadvantage of a high frequency of integration at ectopic sites by illegitimate recombination. The gene targeting efficiency (gene targeting / gene targeting + illegitimate recombination) is 95% for S. cerevisiae, 10-90% for S. pombe, 5-75% for Aspergillus nidulans and 1-30% for Neuropsora crassa using a size of homology of 2-9 Kb (Schaefer, 2001, Current Opinion in Plant Biology 4: 143-150). Especially for filamentous fungi this side effect is quite high, if conventional plasmid vectors are used.

In addition, the efficiency of the gene targeting increases if the length of homologous recombination region is increased (Schaefer, 2001, Current Opinion in Plant Biology 4: 143-150). Thus, plasmid vectors currently used comprise gene fragments of the gene to be knocked out of a size of at least 2000bp as indicated above. The overall size of these plasmid vectors is at least 8000bp (P. J. Punt et. al., 1992, Methods in Enzymology, vol.216, pp 447-457; ; Schaefer, 2001, Current Opinion in Plant Biology 4: 143-150). Since transformation efficiency decreases with the increase of the plasmid vector size, transformation efficiency is unsatisfactury giving rise to long times until positive clones can be identified. This is an obstacle especially to large scale genomic analysis projects or recombinant expression.

Furthermore, currently used plasmid vectors contain many unique restriction sites, causing difficulties in construction of the knock-out (KO-) plasmids and the transformation process. The efficiency of homologous recombination is improved when the KO-plasmid is digested with a restriction enzyme presenting a unique site in the middle of a DNA fragment homologous to the targeted gene. The presence of high amounts of restriction sites especially unique ones in the plasmid backbone decreases the chance of finding a natural restriction site in the appropriate location of the targeted DNA fragment. This problem is usually overcome by modification of the targeted DNA fragment requiring several cloning steps and additional manipulation in terms of molecular biology, which is a disadvantageous and time consuming methology.

Integration of recombinant gen by homolgous recombination in fungi is also a tool to identify gene functions for essential genes: the biochemical characterization of an essential gene cannot be studied by classical knock-out strategy since the mutants carrying a disruption of such a gene are not viable. One way to overexpress such a gene overcomes the problem when a typical phenotype can be assigned to the mutant that overexpresses the gene. Another approach can be to regulate the gene expression by an inducible promoter sequence so that the gene could be expressed or repressed when needed and consequently permits viable mutants to be isolated. As mentioned above, these approaches require at least several thousand bp of the nucleic acid sequence to be studied that need to be integrated in the genome of the fungi together with a plasmid vector comprising the different parts of the nucleic acid sequence. In addition, if the recombinant DNA is integrated at an ectopic site, the identification of the mutant strains becomes more complicated and the position of the integration in the genome may influence the level of expression of the recombinant protein. Taking the aforesaid into consideration,

currently existing plasmid vectors currently used for transformation of filamentous fungi exhibit a lot of disadvantages and are neither suitable for large scale analysis e.g. in functional genomic studies nor convenient for recombinant expression in a filamentous fungi. Additionally, there is a constant need for new selection markers facilitating the selection process.

Thus, object of the present invention was to develop tools for targeted transformation of filamentous fungi that overcome the disadvantages of the state of the art like plasmid vectors suitable for functional genomic studies and recombinant expression and new selection markers.

We have found that the object of the invention is achieved by construction of a plasmid vector for targeted transformation of filamentous fungi comprising

a) an origin of replication for a host organism not originating from the filamentous fungi to be transformed;

b) a selection marker for a host organism not originating from the filamentous fungi to be transformed;

c) a promotor facilitating recombinant expression in filamentous fungi that is functionally linked to the coding region of the hygromycin resistance gene which is functionally linked to a terminator which facilitates transcription termination in filamentous fungi;

wherein the overall size of the elements a), b) and c) does not exceed 4500 bp; and

d) a nucleic acid sequence, which is homologous to nucleic acid sequences of the filamentous fungi to be transformed and makes homologous recombination in the filamentous fungi to be transformed possible.

The term overall size of the elements a), b) and c) designates the combination of the essential elements of the expression vector without the nucleic acid sequence d).

The overall size of the elements a), b) and c) does not exceed 4500 bp, preferaby 4100 bp, more preferably 3700 bp.

In addition to the nucleic acid elements a), b), c) and d), the plasmid vector optionally comprises a cloning site containing rare restriction sites or a TA-cloning site by which further nu-

cleic acid sequences can be cloned easily into the plasmid vector. A TA-cloning site comprises thymidine residues linked onto the 3'-ends of linearized plasmid DNA, which would allow some annealing to occur between the vector and the A-tailed PCR product to be ligated. This process is called TA cloning. Preferably, the vector is modified in such a way that there are only few unique restriction sites left enabling the digestion by commercially available restriction enzymes of the homologous sequence of the targeted gene prior to the transformation.

Filamentous fungi that can be transformed with the vectors of the present invention are non-phytopathogenic filamentous fungi e.g. Neurospora species like Neurospora crassa and phytopathogenic filamentous fungi the phytopathogenic filamentous fungi being preferred. Examples of other non-pythopathogenic filamentous fungi are Aspergillus species such as Aspergillus parasiticus, Aspergillus nidulans, Aspergillus niger and Wagiella such as Wangiella dermatidis. Preferred phytopathogenic filamentous fungi are selected from the group consisting of the genera Alternaria, Podosphaera, Sclerotinia, Physalospora, Botrytis, Corynespora; Colletotrichum; Diplocarpon; Elsinoe; Diaporthe; Sphaerotheca; Cinula, Cercospora; Erysiphe; Sphaerotheca; Leveillula; Mycosphaerella; Phyllactinia; Gloesporium; Gymnosporangium, Leptotthrydium, Podosphaera; Gloedes; Cladosporium; Phomopsis; Phytopora; Phytophthora; Erysiphe; Fusarium; Verticillium; Glomerella; Drechslera; Bipolaris; Personospora; Phaeoisariopsis; Spaceloma; Pseudocercosporella; Pseudoperonospora; Puccinia; Typhula; Pyricularia; Rhizoctonia; Stachosporium; Uncinula; Ustilago; Gaeumannomyces and Fusarium, more preferred from the group consisting of the genera and species Alternaria, Podosphaera, Sclerotinia, Physalospora such as Physalospora canker, Botrytis species such as Botrytis cinerea, Corynespora such as Corynespora melonis; Colletotrichum; Diplocarpon such as Diplocarpon rosae; Elsinoe such as Elsinoe fawcetti, Diaporthe such as Diaporthe citri; Sphaerotheca; Cinula such as Cinula neccata, Cercospora; Erysiphe such as Erysiphe cichoracearum and Erysiphe graminis; Sphaerotheca such as Sphaerotheca fuliginea; Leveillula such as Leveillula taurica; Mycosphaerella; Phyllactinia such as Phyllactinia kakicola; Gloesporium such as Gloesporium kaki; Gymnosporangium such as Gymnosporangium yamadae, Leptotthrydium such as Leptotthrydium pomi, Podosphaera such as Podosphaera leucotricha; Gloedes such as Gloedes pomigena; Cladosporium such as Cladosporium carpophilum; Phomopsis; Phytopora; Phytophthora such as Phytophthora infestans; Verticillium; Glomerella such as Glomerella cingulata; Drechslera; Bipolaris; Personospora; Phaeoisariopsis such as Phaeoisariopsis vitis; Spaceloma such as Spaceloma ampelina; Pseudocercosporella such as Pseudocercosporella herpotrichoides;

Pseudoperonospora; Puccinia; Typhula; Pyricularia such as Pyricularia oryzae; Rhizoctonia; Stachosporium such as Stachosporium nodorum; Uncinula such as Uncinula necator; Ustilago; Gaeumannomyces species such as Gaeumannomyces graminis and Fusarium such as Fusarium dimerium, Fusarium merismoides, Fusarium lateritium, Fusarium decemcellulare, Fusarium poae, Fusarium tricinctum, Fusarium sporotrichioides, Fusarium chlamydosporum, Fusarium moniliforme, Fusarium proliferatum, Fusarium anthophilum, Fusarium subglutinans, Fusarium nygamai, Fusarium oxysporum, Fusarium solani, Fusarium culmorum, Fusarium sambucinum, Fusarium crookwellense, Fusarium avenaceum ssp. avenaceum, Fusarium avenaceum ssp. aywerte, Fusarium avenaceum ssp. nurragi, Fusarium hetrosporum, Fusarium acuminatum ssp. acuminatum, Fusarium acuminatum ssp. armeniacum, Fusarium longipes, Fusarium compactum, Fusarium equiseti, Fusarium scripi, Fusarium polyphialidicum, Fusarium semitectum and Fusarium beomiforme and especially preferred from the genera Fusarium such as Fusarium graminearum, most preferred from the group consisting of the genera and species Fusarium, Fusarium dimerium, Fusarium merismoides, Fusarium lateritium, Fusarium decemcellulare, Fusarium poae, Fusarium tricinctum, Fusarium sporotrichioides, Fusarium chlamydosporum, Fusarium moniliforme, Fusarium proliferatum, Fusarium anthophilum, Fusarium subglutinans, Fusarium nygamai, Fusarium oxysporum, Fusarium solani, Fusarium culmorum, Fusarium sambucinum, Fusarium crookwellense, Fusarium avenaceum ssp. avenaceum, Fusarium avenaceum ssp. aywerte, Fusarium avenaceum ssp. nurragi, Fusarium hetrosporum, Fusarium acuminatum ssp. acuminatum, Fusarium acuminatum ssp. armeniacum, Fusarium longipes, Fusarium compactum, Fusarium equiseti, Fusarium scripi, Fusarium polyphialidicum, Fusarium semitectum and Fusarium beomiforme wherein Fusarium graminearum is most preferred.

The host organism in which the origin of replication a) is functionally active essentially serves to construct and propagate the plasmid vector of the invention. The host organism must be genetically different from the filamentous fungi to be transformed, since replication of the plasmid vector should not take place in the filamentous fungi to be transformed but is desired in the host organism, due to the use of the origin of replication a). Host organisms which may be used are all common microorganisms which can easily be manipulated by genetic engineering. Preferred host organisms are Gram-negative bacteria such as the genera Escherichia and Salmonella e.g. Escherichia coli and Salmonella thyplimurium or Gram-positive bacteria such as the genera Bacillus and Streptomyces, e.g. Bacillus subtilis and Streptomyces nidulans. Particularly preferred are gram-negative bacteria such as

Escherichia, e.g. Escherichia coli.

Preferred origins of replication (ori) are the col E1 ori, the f1 ori.

The term "selection marker for a host organism" set forth in b) means a gene or the expression product of the gene. Preferred meanings are genes whose expression causes resistance of the host organism to antibiotics, by preference resistance to kanamycin, chloramphenicol, tetracycline, zeocin or ampicillin, and particularly preferred ampicillin and kanamycin.

In a preferred embodiment, the element a) of the plasmid vector according to the invention comprises a col E1 origin of replication and the ampicillin resistance gene as selection marker for the host organism.

The element c) is hereinbelow termed "hygromycin cassette". The coding region of the hygromycin resistance gene (hereinbelow termed "hygromycin gene") is known by the skilled artisan (Gritz L. and Davies J. ,1983, Gene 25, 179-188, Kaster, K.R., Burgett S.G. and Ingolia T.D., 1984, Curr. Genet. 8,353-358) and has a length of 1026bp.

Examples of suitable promotors to which the coding region of the hygromycin gene is functionally linked, are the GPD-1-, PX6-, TEF-, CUP1-, PGK-, GAP1-, TPI, PHO5-, AOX1, GAL10/CYC-1, CYC1, OliC-, ADH-, TDH-, Kex2-, MFα-, or the NMT-promotor (Degryse et al., Yeast 1995 Jun 15; 11(7):629-40; Romanos et al. Yeast 1992 Jun;8(6):423-88; Benito et al. Eur. J. Plant Pathol. 104, 207-220 (1998); Cregg et al. Biotechnology (N Y) 1993 Aug;11(8):905-10; Luo X., Gene 1995 Sep 22;163(1):127-31; Nacken et al., Gene 1996 Oct 10;175(1-2):253-60; Turgeon et al., Mol Cell Biol 1987 Sep;7(9):3297-305), preferably the CYC1-, ADH-, TDH-, Kex2-, GPD-1-, PX6, TEF-, CUP1-, PGK-, GAP1-, TPI, PHO5- or AOX1-promotor, more preferably the GPD-1-, PX6, TEF- or the CUP1-promotor, most preferably the GPD1 or the TEF-promotor.

Examples of suitable terminators that are functionally linked to the coding region of the hygromycin gene are the AOX1-, nos-, PGK-, TrpC- or the CYC1-terminator (Degryse et al., Yeast 1995 Jun 15; 11(7):629-40; Brunelli et al. Yeast 1993 Dec9(12): 1309-18; Frisch et al., Plant Mol. Biol. 27 (2), 405-409 (1995); Scorer et al., Biotechnology (N.Y.) 12 (2), 181-184 (1994), Genbank acc. number Z46232; Punt et al., (1987) Gene 56 (1),

117-124)), preferably the CYC1- or nos-terminator, more preferably the nos-terminator.

In a preferred embodiment, the hygromycin cassette comprises a GPD-1 promotor functionally linked to the coding region of the hygromycin region which is functionally linked to the nos-terminator.

A functional linkage is understood to mean the sequential arrangement of promoter and coding sequence, of coding sequence and terminator or of promoter, coding sequence and terminator in such a manner that each of the regulatory elements can, upon expression of the coding sequence, fulfil its function for the recombinant expression of the nucleic acid sequence. Direct linkage in the chemical sense is not necessarily required for this purpose. Preferred arrangements are those in which the hygromycin gene to be expressed recombinantly is positioned downstream of the sequence which acts as promoter, so that the two sequences are linked covalently to each other. The distance between the promoter sequence and the nucleic acid sequence to be expressed recombinantly is preferably less than 100 base pairs, especially preferably less than 50 base pairs, and very especially preferably less than 10 base pairs. The distance between the terminator sequence and the nucleic acid sequence to be expressed recombinantly is preferably less than 100 base pairs, especially preferably less than 50 base pairs, and very especially preferably less than 10 base pairs. However, further sequences which, for example, exert the function of a linker with certain restriction enzyme cleavage sites, or of a signal peptide, may also be positioned between the two sequences.

These vectors are not only much more smaller than the currently used plasmid vectors, but also exhibit a high transformation efficiency. Surprisingly, a high transformation efficiency can be gained even if small DNA-fragments of at least 300bp, preferably at least 400bp, more preferably at least 450bp, most preferably at least 500bp of the nucleic acid sequence d) to be analyzed are used. The average degree of illegitimate recombination is below 30%, preferably below 25%, more preferably 20% and most preferably between 0 and 15%.

The nucleic acid sequence d) has a homology of at least 80% to the nucleic acid sequence of the filamentous fungi to be transformed, preferably at least 90%, more preferably at least 95% and most preferably at least 100%.

In a preferred embodiment, the nucleic acid sequence d) originates from a filamentous fungi and has a length of at least 300bp, preferably 400bp, more preferably at least 450bp, and most preferably at least 500bp. These lengths are suitable for functional genomic studies for which a high number of transformants is required. Also nucleic acid sequences exceeding 500bp can be used, e.g. for the purpose of recombinant expression.

If the nucleic acid sequence d) is to be expressed recombinantly in the filamentous fungi, it can be functionally linked to a promotor e) and optionally to a terminator f).

Examples of suitable promotors e) are for example the AUG1-, GPD-1-, PX6-, TEF-, CUP1-, PGK-, GAP1-, TPI, PHO5-, AOX1, GAL10/CYC-1, CYC1, OliC-, ADH-, TDH-, Kex2-, MFα- or the NMT-promotor or combinations of the aforementioned promotors (Degryse et al., Yeast 1995 Jun 15; 11(7):629-40; Romanos et al. Yeast 1992 Jun;8(6):423-88; Benito et al. Eur. J. Plant Pathol. 104, 207-220 (1998); Cregg et al. Biotechnology (N Y) 1993 Aug;11(8):905-10; Luo X., Gene 1995 Sep 22;163(1):127-31; Nacken et al., Gene 1996 Oct 10;175(1-2): 253-60; Turgeon et al., Mol Cell Biol 1987 Sep;7(9):3297-305).

Examples of suitable terminators f) are the NMT-, Gcy1-, TrpC-, AOX1-, nos-, the PGK- or the CYC1-terminator (Degryse et al., Yeast 1995 Jun 15; 11(7):629-40; Brunelli et al. Yeast 1993 Dec9(12): 1309-18; Frisch et al., Plant Mol. Biol. 27 (2), 405-409 (1995); Scorer et al., Biotechnology (N.Y.) 12 (2), 181-184 (1994), Genbank acc. number Z46232; Zhao et al. Genbank acc number : AF049064; Punt et al., (1987) Gene 56 (1), 117-124).

The nucleic acid sequence d) can be also functionally linked to an affinity tag to purify the encoded protein and/or to a reporter gene to study biochemical properties of the nucleic acid sequence d) in vivo, respectively.

"Reporter genes" encode readily quantifiable proteins. Using these genes, an assessment of transformation efficacy or of the site or time of expression can be made via growth, fluorescence, chemoluminescence, bioluminescence or resistance assay or via photometric measurement (intrinsic color) or enzyme activity. Very especially preferred in this context are reporter proteins (Schenborn E, Groskreutz D. Mol. Biotechnol. 1999; 13(1):29-44) such as the "green fluorescence protein" (GFP) (Gerdes HH and Kaether C, FEBS Lett. 1996; 389(1):44-47; Chui WL et al., Curr. Biol. 1996, 6:325-330; Leffel SM et al., Biotechniques.

23(5):912-8, 1997), chloramphenicol acetyl transferase, a luciferase (Giacomin, Plant Sci. 1996, 116:59-72; Scikantha, J. Bact. 1996, 178:121; Millar et al., Plant Mol. Biol. Rep. 1992 10:324-414), and luciferase genes, in general β-galactosidase or β-glucuronidase (Jefferson et al., EMBO J. 1987, 6, 3901-3907), the Ura3 gene, the Ilv2 gene, the 2-desoxyglucose-6-phosphate phosphatase gene, β-lactamase gene, the neomycin phosphotransferase gene, the hygromycin phosphotransferase gene, or the BASTA (= gluphosinate) resistance gene.

The term "affinity tag" denotes a peptide or polypeptide whose coding nucleic acid sequence can be fused to the nucleic acid sequence d) either directly or using a linker, by classical cloning techniques. The affinity tag serves to isolate the recombinant target protein by means of affinity chromatography. The abovementioned linker can optionally comprise a protease cleavage site (for example for thrombin or factor Xa), whereby the affinity tag can be cleaved off from the target protein, as required. Examples of customary affinity tags are the "his-tag", for example from Quiagen, Hilden, "strep-tag", "myc-tag" (Invitrogen, Carlsberg), New England Biolab's tag which consists of a chitin binding domain and an intein, and what is known as the CBD-tag from Novagen.

In a particularly preferred embodiment, the plasmid vector comprises an coli E1 ori, the ampicillin resistance gene as selection marker, a GPD-1 promotor functionally linked to the coding region of the hygromycin resistance gene which is functionally linked to the nos-terminator.

Preferably, the vector also comprises a multiple cloning site comprising an appropriate restriction enzyme site. Appropriate restriction sites are well known by the skilled artisan.

In a further preferred embodiment, the plasmid vector additionally comprises a TA-cloning site to facilitate the overall cloning procedure.

Examples of particularly preferred embodiments are set forth in Fig. 1 and 2.

All of the above mentioned embodiments of plasmid vectors are hereinbelow termed "plasmid vector (or vector) according to the invention".

A vector according to the invention may also comprise at least one additional selection marker.

If a plasmid is used for recombinant expression in host organisms, a marker is required indicating the sucessful transfer of the plasmid vector DNA into the filamentous fungi to be transformed.

Surprisingly, we have found that the gene fragments of the polyketide synthase are a my suitable selection marker. The term "selection marker" referring to the polyketide synthase herein means a nucleic acid sequence.

More precisely, the term "selectable marker", "selection marker" or "marker" used in connection with polyketide synthetase for transformation of filamentous fungi means a nucleic acid sequence encoding a polyketide synthetase or fragments of the aforementioned nucleic acid sequence. Preferred embodiments of the aforementioned marker as well as preferred embodiments of methods of use of the respective marker are described herein below.

Polyketide synthases are multifunctional enzymes that are involved in the biosynthesis of several important polyketides. Polyketides constitute a large and highly diverse group of secondary metabolites, synthesized by bacteria, fungi and plants and algae. They include antibiotics, compounds with mycotoxic activity, and compounds within pigment biosynthetic pathways. Furthermore a polyketide synthase is described to be required for fungal virulence of Cochliobolus heterostrophous toward maize (Yang et al., 1996 PMID:8953776). Polyketide Synthetases are furthermore known from Wangiella dermatidis (pubMedID:11179356), from Aspergillus nidulans (Swiss-prot ID: Q03149) and from Aspergillus parasiticus (Swiss-Prot ID:Q12053).

The use of polyketide synthase as selectable marker for recombinant expression in filamentous fungi has not yet been described.

The present invention also encompasses a selection marker comprising a nucleic acid sequence encoding a polyketide synthetase fragment, wherein said nucleic acid sequence comprises

i. a nucleic acid sequence shown in SEQ ID NO:1 or SEQ ID NO:2; or

ii. parts of the nucleic acid sequence as defined in i. consisting of at least 300bp.

Parts or segments of nucleic acid sequences set forth in ii. consist of at least 300bp, preferably at least 400bp, more preferably at least 450bp, most preferably at least 500bp of the nucleic acid sequences. In a further preferred embodiment, those parts are selected from SEQ ID NO:1, preferably from 732bp to 5881bp of SEQ ID NO:1 e.g. from 2236bp to 2870bp.

Furthermore, the present invention encompasses a selection marker comprising a nucleic acid sequence encoding a polyketide synthetase or a polyketide synthetase fragment, wherein said nucleic acid sequence comprises

i. a nucleic acid sequence shown in SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5; or

ii. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:6 by back translation; or

iii. a functional equivalent of the nucleic acid sequence set forth in i) which is encoded by an amino acid sequence that has at least an identity of 50% with the SEQ ID NO:6; or

iv. parts of the nucleic acid sequence as defined in i., ii. or iii. consisting of at least 300bp.

v. parts of the nucleic acid sequence as defined in i., ii. or iii. consisting of at least 300bp comprising

a) a nucleic acid sequence shown in SEQ ID NO:7; or

b) a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:8 by back translation; or

c) a functional equivalent of a nucleic acid sequence set forth in a) which is encoded by an amino acid sequence that has at least an identity of 85% with the SEQ ID NO:8.

Parts or segments of nucleic acid sequences set forth in iii. or v. consist of at least 300bp, preferably at least 400bp, more preferably at least 450bp, most preferably at least 500bp of the nucleic acid sequences. Preferably, the aforementioned parts or segments of nucleic acid sequences are those set forth in v.a), v.b) or v.c), more preferably those set forth in v.a) or v.b) and most preferably those set forth in v.a). For example, those parts

can be selected from 2234bp to 2865bp of SEQ ID NO:3.

The functional equivalents of the nucleic acid sequence set forth in iv. are encoded by an amino acid sequence that has at least an identity of 50%, 51%, 52%, 53%, 54%, 55%, 56%, 57%, 58%, 59%, 60%, 61%, 62%, 63%, 64% or 65% or preferably of 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78% or 79% more preferably of 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89% or 90% most preferably of 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98% or 99% with SEQ ID NO:6.

The functional equivalents of the nucleic acid sequence set forth in v.c) are encoded by an amino acid sequence that has at least an identity of 85%, 86%, 87% or 88% or preferably of 89%, 90%, 91%, 92% or 93% more preferably of 94%, 95% or 96% most preferably of 97%, 98% or 99% with SEQ ID NO:8.

Preferred are nucleic acid sequences as defined above originating from filamentous fungi, preferably phytopathogenic filamentous fungi selected from the group consisting of the genera Neurospora, Alternaria, Podosphaera, Sclerotinia, Physalospora, Botrytis, Corynespora; Colletotrichum; Diplocarpon; Elsinoe; Diaporthe; Sphaerotheca; Cinula, Cercospora; Erysiphe; Sphaerotheca; Leveillula; Mycosphaerella; Phyllactinia; Gloesporium; Gymnosporangium, Leptotthrydium, Podosphaera; Gloedes; Cladosporium; Phomopsis; Phytopora; Phytophthora; Erysiphe; Fusarium; Verticillium; Glomerella; Drechslera; Bipolaris; Personospora; Phaeoisariopsis; Spaceloma; Pseudocercosporella; Pseudoperonospora; Puccinia; Typhula; Pyricularia; Rhizoctonia; Stachosporium; Uncinula; Ustilago; Gaeumannomyces and Fusarium, more preferred from the group consisting of the genera and species Alternaria, Podosphaera, Sclerotinia, Physalospora such as Physalospora canker, Botrytis species such as Botrytis cinerea, Corynespora such as Corynespora melonis; Colletotrichum; Diplocarpon such as Diplocarpon rosae; Elsinoe such as Elsinoe fawcetti, Diaporthe such as Diaporthe citri; Sphaerotheca; Cinula such as Cinula neccata, Cercospora; Erysiphe such as Erysiphe cichoracearum and Erysiphe graminis; Sphaerotheca such as Sphaerotheca fuliginea; Leveillula such as Leveillula taurica; Mycosphaerella; Phyllactinia such as Phyllactinia kakicola; Gloesporium such as Gloesporium kaki; Gymnosporangium such as Gymnosporangium yamadae, Leptotthrydium such as Leptotthrydium pomi, Podosphaera such as Podosphaera leucotricha; Gloedes such as Gloedes pomigena; Cladosporium such as Cladosporium carpophilum; Phomopsis; Phytopora; Phytophthora such as Phytophthora infestans; Verticillium; Glomerella such as Glomerella cingulata; Drechslera; Bipolaris; Personospora; Phaeoisariopsis such as Phaeoisariopsis vitis; Spaceloma such as Space-

loma ampelina; Pseudocercosporella such as Pseudocercosporella herpotrichoides; Pseudoperonospora; Puccinia; Typhula; Pyricularia such as Pyricularia oryzae; Rhizoctonia; Stachosporium such as Stachosporium nodorum; Uncinula such as Uncinula necator; Ustilago; Gaeumannomyces species such as Gaeumannomyces graminis and Fusarium such as Fusarium dimerium, Fusarium merismoides, Fusarium lateritium, Fusarium decemcellulare, Fusarium poae, Fusarium tricinctum, Fusarium sporotrichioides, Fusarium chlamydosporum, Fusarium moniliforme, Fusarium proliferatum, Fusarium anthophilum, Fusarium subglutinans, Fusarium nygamai, Fusarium oxysporum, Fusarium solani, Fusarium culmorum, Fusarium sambucinum, Fusarium crookwellense, Fusarium avenaceum ssp. avenaceum, Fusarium avenaceum ssp. aywerte, Fusarium avenaceum ssp. nurragi, Fusarium hetrosporum, Fusarium acuminatum ssp. acuminatum, Fusarium acuminatum ssp. armeniacum, Fusarium longipes, Fusarium compactum, Fusarium equiseti, Fusarium scripi, Fusarium polyphialidicum, Fusarium semitectum and Fusarium beomiforme and especially preferred from the genera Fusarium such as Fusarium graminearum, most preferred from the group consisting of the genera and species Fusarium, Fusarium dimerium, Fusarium merismoides, Fusarium lateritium, Fusarium decemcellulare, Fusarium poae, Fusarium tricinctum, Fusarium sporotrichioides, Fusarium chlamydosporum, Fusarium moniliforme, Fusarium proliferatum, Fusarium anthophilum, Fusarium subglutinans, Fusarium nygamai, Fusarium oxysporum, Fusarium solani, Fusarium culmorum, Fusarium sambucinum, Fusarium crookwellense, Fusarium avenaceum ssp. avenaceum, Fusarium avenaceum ssp. aywerte, Fusarium avenaceum ssp. nurragi, Fusarium hetrosporum, Fusarium acuminatum ssp. acuminatum, Fusarium acuminatum ssp. armeniacum, Fusarium longipes, Fusarium compactum, Fusarium equiseti, Fusarium scripi, Fusarium polyphialidicum, Fusarium semitectum and Fusarium beomiforme, wherein Fusarium graminearum is most preferred.

Preferred non-phytopathogenic filamentous fungi are fungi of the group consisting of the genera Neurospora such as Neurospora crassa, Aspergillus such as Aspergillus parasiticus, Aspergillus nidulans, Aspergillus niger and Wangiella such as Wangiella dermatidis.

The term "comprising" means that the nucleic acid sequence according to the invention can be flanked by additional nucleic acid sequences that have on the 5' end a sequence length of at least 1000 bp and preferably at least 500 bp, more preferably at least 100bp, most preferably at least 50bp and on the 3' a sequence length of at least 1000 bp and preferably at least 500 bp,

more preferably at least 100 bp and most preferably at least 50bp.

"Functional equivalents" in the present context describe nucleic acid sequences which hybridize under standard conditions with the nucleic acid sequence or portions of the nucleic acid sequence having the function of the a selection marker.

It is advantageous to use short oligonucleotides of a length of 10-50 bp, preferably 15-40 bp, for example of the conserved or other regions, which can be determined via comparisons with other related genes in a manner known to the skilled worker for the hybridization. Alternatively, it is also possible to use longer fragments of the nucleic acids according to the invention or the complete sequences for the hybridization. These standard conditions vary depending on the nucleic acid used, viz. oligonucleotide, longer fragment or complete sequence, or depending on which type of nucleic acid, viz. DNA or RNA, is being used for the hybridization. Thus, for example, the melting temperatures for DNA:DNA hybrids are approx. 10°C lower than those of DNA:RNA hybrids of equal length.

Standard conditions are understood to mean, depending on the nucleic acid, for example temperatures between 42 and 58°C in an aqueous buffer solution with a concentration of between 0.1 and 5 x SSC (1 x SSC = 0.15 M NaCl, 15 mM sodium citrate, pH 7.2) or additionally in the presence of 50% formamide such as, for example, 42°C in 5 x SSC, 50% formamide. The hybridization conditions for DNA:DNA hybrids are advantageously 0.1 x SSC and temperatures of between approximately 20°C and 45°C, preferably between approximately 30°C and 45°C. The hybridization conditions for DNA:RNA hybrids are advantageously 0.1 x SSC and temperatures of between approximately 30°C and 55°C, preferably between approximately 45°C and 55°C. These temperatures stated for the hybridization are melting temperature values which have been calculated by way of example for a nucleic acid with a length of approx. 100 nucleotides and a G + C content of 50% in the absence of formamide. The experimental conditions for DNA hybridization are described in specialist textbooks of genetics such as, for example, Sambrook et al., "Molecular Cloning", Cold Spring Harbor Laboratory, 1989 and can be calculated using formulae known to the skilled worker, for example as a function of the length of the nucleic acids, the type of the hybrids or the G + C content. The skilled worker can find more information on hybridization in the following textbooks: Ausubel et al. (eds), 1985, Current Protocols in Molecular Biology, John Wiley & Sons, New York; Hames and Higgins (eds),

1985, Nucleic Acids Hybridization: A Practical Approach, IRL Press at Oxford University Press, Oxford; Brown (ed), 1991, Essential Molecular Biology: A Practical Approach, IRL Press at Oxford University Press, Oxford.

A functional equivalent is furthermore also understood to mean, in particular, natural or artificial mutations of the relevant nucleic acid sequences of the polyketide synthetase (PKS) as set forth in SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5 and its homologs from other organisms , wherein mutations comprise substitutions, additions, deletions, inversions or insertions of one or more nucleotide residues. This may also lead to a modification of the corresponding amino acid sequence of the PKS by substitution, insertion or deletion of one or more amino acids.

Thus, the scope of the present invention also extends to, for example, those nucleotide sequences which are obtained by modification of the nucleic acid sequence of the selection marker described by SEQ ID NO:1 or by SEQ ID NO:2 or SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5 respectively. The purpose of such a modification can be, for example, the insertion of further cleavage sites for restriction enzymes, the removal of excess DNA, or the addition of further sequences. Said nucleic acid sequences should still maintain the desired function as marker for targeted transformation, despite the deviating nucleic acid sequence.

The term "identity" or "homology" between two nucleic acid sequences or polypeptide sequences is defined by the identity of the nucleic acid sequence/polypeptide sequence by in each case the entire sequence length, which is calculated by alignment with the aid of the program algorithm GAP (Wisconsin Package Version 10.0, University of Wisconsin, Genetics Computer Group (GCG), Madison, USA), setting the following parameters:

Gap Weight: 8 Length Weight: 4

Average Match: 2,912 Average Mismatch:-2,003

The term homology when used herein is the same as the term identity.

Functional equivalents thus encompass naturally occurring variants of the sequences described herein, and also artificial, for example chemically synthesized, nucleic acid sequences adapted to

the codon usage, or the amino acid sequences derived therefrom.

Moreover, SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5, nucleic acid sequences derived from the amino acid sequence SEQ ID NO:6 by back translation or parts of the aforementioned nucleic acid sequences can be used for the detection and isolation of functional equivalents of other fungi on the basis of sequence identities. In this context, part or all of the sequence of the SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5 or nucleic acid sequences derived from the amino acid sequence SEQ ID NO:6 by back translation can be used as probe (e.g. hybridization probe) for screening in a genomic library or a cDNA library of the fungal species in question or in a computer search for sequences of functional equivalents in electronic databases. Especially for computer search for sequences of functional equivalents in electronic databases, the amino acid sequence SEQ ID NO:6 or parts of the amino acid sequence SEQ ID NO:6 are useful.

For the preparation of hybridization probes, SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5 or parts of the aforementioned nucleic acid sequences can be used. The preparation of these probes and the experimental procedure are known. For example, this can be effected via the tailor-made preparation of radioactive or nonradioactive probes by means of PCR and the use of suitably labeled oligonucleotides, followed by hybridization experiments. The technologies required for this purpose are given, for example, in T. Maniatis, E.F. Fritsch and J. Sambrook, "Molecular Cloning: A Laboratory Manual", Cold Spring Harbor Laboratory, Cold Spring Harbor, NY (1989). The probes in question can furthermore be modified by standard technology (lit. SDM or random mutagenesis) in such a way that they can be employed for other purposes, for example as probe which hybridizes specifically with mRNA and the corresponding coding sequences in order to analyze the corresponding sequences in other organisms.

Furthermore, the cDNA could be used to engineer recombinant microorganisms to produce polyketide agents of pharmaceutical or agricultural interest as described by Pfeifer et al. (Pfeifer BA, Admiraal SJ, Gramajo H, Cane DE, Khosla C., Science 2001 Mar 2;291(5509):1790-2). Thus, the present invention also comprises polypeptides with the biological acitivity of a polyketide synthetase encoded by a nucleic acid sequence comprising

i. a nucleic acid sequence shown in SEQ ID NO:5 or

ii. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:6 by back translation; or

iii. nucleic acid sequence which is encoded by a functional analogue of an amino acid sequence that has at least an identity of 50% with the SEQ ID NO:6.

The term "functional analogues" describes nucleic acid sequences which are capable of bringing about the expression, in a filamentous fungi, of a polypeptide with the biological activity of polyketide synthetase and which can be deduced from an amino acid sequence by back translation which has a defiened degree of identity with SEQ ID NO:6. The functional analogues set forth in iii) have at least an identity of 50%, 51%, 52%, 53%, 54%, 55%, 56%, 57%, 58%, 59%, 60%, 61%, 62%, 63%, 64% or 65% or preferably of 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78% or 79% more preferably of 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89% or 90% most preferably of 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98% or 99% with the SEQ ID NO:6.

Thus, the present invention also encompasses, for example, those nucleotide sequences which are obtained by modification of the abovementioned nucleic acid sequences. For example, such modifications can be generated by techniques with which the skilled worker is familiar, such as "site directed mutagenesis", "error prone PCR", "DNA shuffling" (Nature 370, 1994, pp.389-391) or "staggered extension process" (Nature Biotechnol. 16, 1998, pp.258-261). The purpose of such a modification can be, for example, the insertion of further cleavage sites for restriction enzymes, the removal of DNA in order to truncate the sequence, the substitution of nucleotides in order to optimize the codons, or the addition of further sequences. Proteins which are encoded via modified nucleic acid sequences must retain the desired functions despite a deviating nucleic acid sequence.

Functional analogues thus comprise naturally occuring variants of the herein-described sequences and artificial nucleic acid sequences, for example those which have been obtained by chemical synthesis and which are adapted to the codon usage, and also the amino acid sequences derived from them.

As explained above, also the expression cassette or the vector comprising a PKS encoding nucleic acid sequence may comprise at least an additional selection marker, preferably the hygromycin resistance gene so that in a particular preferred embodiment, the selection of the succesfully transformed filamentous fungi can be

carried out by hygromycin resistance of successfully transformed clones and by the presence of pigment (color) of successfully transformed clones. Most preferably, the vector comprising the PKS encoding nucleic acid sequence is a vector according to the invention comprising a PKS encoding nucleic acid sequence. In addition to the aforementioned selection method homologous recombination can be confirmed by PCR based on oligonucleotides preferably derived from the vector sequence flanking the 5 and 3 region of the gene to be inserted. Specific examples of these primers are given in the examples.

The invention furthermore relates to the use of polyketide synthetase encoding nucleic acid sequences as marker for targeted transformation in filamentous fungi.

Preferably, the present invention comprises the use of a nucleic acid sequence comprising

a) a nucleic acid sequence encoding a polyketide synthetase; or

b) parts of the nucleic acid sequence as defined in i., ii. or iii. consisting of at least 300bp

for transformation of filamentous fungi.

Preferred is the use of a nucleic acid sequence as marker for targeted transformation in filamentous fungi said nucleic acid comprising

i. a nucleic acid sequence shown in SEQ ID NO:1 or SEQ ID NO:2; or

ii. parts of the nucleic acid sequence as defined in i. consisting of at least 300bp, preferably at least 400bp, more preferably at least 450bp and most preferably at least 500bp;

Equally preferred is the use of a nucleic acid sequence as marker for targeted transformation in filamentous fungi, said nucleic acid comprising

iii. a nucleic acid sequence shown in SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5; or

iv. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:6 by back translation; or

v. a functional equivalent of the nucleic acid sequence set forth in i) or iii) which is encoded by an amino acid sequence that has at least an identity of 40% with the SEQ ID NO:6; or

vi. a nucleic acid sequence shown in SEQ ID NO:9 or SEQ ID NO: 11;

vii. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:10, SEQ ID NO:12 or SEQ ID NO:13 by back translation; or

viii. parts of the nucleic acid sequence as defined in iii., iv, v., vi or vii. consisting of at least 300bp, preferably at least 400bp, more preferably at least 450bp and most preferably at least 500bp; or

ix. parts of the nucleic acid sequence as defined in iii., iv, v., vi or vii. consisting of at least 300bp, preferably at least 400bp, more preferably at least 450bp, and most preferably at least 500bp comprising

a) a nucleic acid sequence shown in SEQ ID NO:7; or

b) a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:8 by back translation; or

c) a functional equivalent of a nucleic acid sequence set forth in a) which is encoded by an amino acid sequence that has at least an identity of 68% with the SEQ ID NO:8.

The nucleic acid sequences according to i. to ix encode for a polypeptide with the biological function of a polyketide synthetase or for a fragment of the aforementioned polypeptide.

Under the aforementioned sequences, the nucleic acid sequences according to i., ii., iii., iv., v. as well as parts of the aforementioned nucleic acid sequence consisting of at least 300bp, preferably at least 400bp, more preferably at least 450bp, most preferably at least 500bp are preferred. Those parts are preferably those set forth in ix.

Preferred phytopathogenic and non-phytopathogenic filamentous fungi are those mentioned above. The aforementioned nucleic acid sequences are hereinbelow also termed "PKS marker". Preferably, the term "PKS marker" designates nucleic acid sequences according to i., ii., iii., iv., v. as well as parts of the aforementioned nucleic acid sequence consisting of at least 300bp, preferably at least 400bp, more preferably at least 450bp, and most preferably at least 500bp are preferred.

The functional equivalents of the nucleic acid sequence set forth in iv. can be deduced from a functional equivalent of the amino acid sequence shown in SEQ ID NO:6 by back translation having at least an identity of 40%, 41%, 42%, 43%, 44%, 45%, 46%, 47%, 48% or 49% preferably of 50%, 51%, 52%, 53%, 54%, 55%, 56%, 57%, 58%, 59%, 60%, 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78% or 79% more preferably of 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89% or 90% and most preferably of 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98% or 99% with the SEQ ID NO:6.

The functional equivalents of the nucleic acid sequence set forth in ix.c) can be deduced from a functional equivalent of the amino acid sequence shown in SEQ ID NO:8 by back translation having at least an identity of 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77% or 78% preferably of 79%, 80%, 81%, 82%, 83%, 84% or 85% more preferably of 86%, 87%, 88%, 89% or 90% most preferably of 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98% or 99% with the SEQ ID NO:8.

The use of a PKS marker for targeted transformation of filamentous fungi can be based on significant reduction in the amount of polyketide synthetase which is present in a filamentous fungi. A reduction in the amount the polyketide synthetase means that the amount of polypeptide is reduced via recombinant methods.

Reduction via recombinant methods can involve "antisense techniques", which describes a technology for the suppression (reduction) of expression of polyketide synthetase, where a PKS marker is transformed into the respective filamentous fungi in "antisense" orientation under the control of a suitable promoter. This method is used preferably for Aspergillus species, and more preferrably for Aspergillus nidulans. The technologies required herefore are well known by the skilled artisan (for example see Bautista et al., Appl. Environ. Microbiol. 2000; 66(10) 4579-81). Suitable vectors therefore comprise an expression cassette comprising

a) a promotor sequence in functional linkage with a PKS marker in antisense orientation; and optionally

b) further genetic control sequences functionally linked to a nucleic acid sequence according to a).

The afore-mentioned expression cassette is hereinbelow termed as "PKS Marker expression cassette".

The term "expression cassette" can be defined as follows: An expression cassette comprises a nucleic acid sequence which should be expressed, linked functionally to at least one genetic control element, such as a promoter, and, advantageously, a further control element, such as a terminator. Examples of suitable promotors and terminators are given above. The nucleic acid sequence of the expression casette can be, for example, a genomic or complementary DNA sequence or an RNA sequence, and the semisynthetic or fully synthetic analogs thereof. These sequences can exist in linear or circular form, extrachromosomally or integrated into the genome. The nucleic acid sequences in question can be synthesized or obtained naturally or comprise a mixture of synthetic and natural DNA components, and consist of a variety of heterologous gene segments from various organisms.

Artificial nucleic acid sequences are also suitable in this context as long as they make possible the expression, in a cell or organism, of a polypeptide encoded by a nucleic acid sequence according to the invention and having the biological activity of a polyketide synthetase. For example, synthetic nucleotide sequences can be generated which have been optimized with regard to the codon usage of the organisms to be transformed.

All of the abovementioned nucleotide sequences can be generated from the nucleotide units by chemical synthesis in the manner known per se, for example by fragment condensation of individual, overlapping complementary nucleotide units of the double helix. Oligonucleotides can be synthesized chemically for example in the manner known per se using the phosphoamidite method (Voet, Voet, 2nd Edition, Wiley Press New York, pp. 896-897). When preparing an expression cassette, various DNA fragments can be manipulated in such a way that a nucleotide sequence with the correct direction of reading and the correct reading frame is obtained. The nucleic acid fragments are linked to each other via general cloning techniques as are described, for example, in T. Maniatis, E.F. Fritsch and J. Sambrook, "Molecular Cloning: A Laboratory Manual", Cold Spring Harbor Laboratory, Cold Spring Harbor, NY (1989) and in T.J. Silhavy, M.L. Berman and L.W. Enquist, Experi-

ments with Gene Fusions, Cold Spring Harbor Laboratory, Cold Spring Harbor, NY (1984) and in Ausubel, F.M. et al., "Current Protocols in Molecular Biology", Greene Publishing Assoc. and Wiley-Interscience (1994).

The term "genetic control element" describes sequences which have an effect on the transcription and, if appropriate, translation of the nucleic acids according to the invention in prokaryotic or eukaryotic organisms. Examples are terminators. Examples of suitable terminators are given above. In addition to the afore-mentioned control sequences, or instead of these sequences, the natural regulation of these sequences may still be present before the actual structural genes and may, if appropriate, have been modified genetically in such a way that the natural regulation has been switched off and the expression of the target gene has been modified, that is to say increased or reduced. The choice of the control sequence depends on the host organism or starting organism. Genetic control sequences furthermore also comprise the 5'-untranslated region, introns or the noncoding 3' region of genes. Control sequences are furthermore understood as meaning those which make possible a homologous recombination or insertion into the genome of a host organism or which permit the removal from the genome. Genetic control sequences also comprise further promoters, promoter elements or minimal promoters.

The transcription of the PKS marker leads to suppression of the transcription of the natural polyketide synnthethase gene, which can be detected by loss of color of the transformed fungi relative to the respective wild-type strain.

In a preferred embodiment, the reduction via recombinant methods is based on a gene knock out of the polyketide synthethase gene using either an expression cassette additionally comprising the PKS marker or a vector comprising the PKS marker in the respective filamentous fungi. Disruption of the PKS marker will lead to a loss of color.

Preferred phytopathogenic filamentous fungi are those mentioned above. Preferred non-phytopathogenic filamentous fungi are fungi of the group consisting of the genera Aspergillus such as Aspergillus parasiticus, Aspergillus nidulans and Wangiella such as Wangiella dermatidis.

In this connection, the selection of the functional equivalent for the use as marker gene depends on the fungi to be transformed. By preference, the polyketide synthetase fragment has an identity of at least 80%, preferably at least 81%, 82%, 83%, 84%,

85%, 86%, 87%, 88%, 89%, 90%, and especially preferably at least 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% identity with the polyketide synthetase of the fungi to be transformed.

For example, for transformation of Fusarium graminearum, a nucleic acid sequence can be selected comprising

i. a nucleic acid sequence shown in SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5; or

ii. a nucleic acid sequence that has at least an identity of 80% SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5; or

iii. parts of the nucleic acid sequence as defined in i. or ii. consisting of at least 300bp, preferably at least 400bp, more preferably at least 450bp, and most preferably at least 500bp.

iv. parts of the nucleic acid sequence as defined in i. or ii. consisting of at least 300bp, preferably at least 400bp, more preferably at least 450bp, and most preferably at least 500bp comprising

a) a nucleic acid sequence shown in SEQ ID NO:7; or

c) a nucleic acid sequence that has at least an identity of 80% with the SEQ ID NO:8.

As mentioned above, another embodiment of the present invention is plasmid vectors for targeted transformation of filamentous fungi comprising a PKS marker. These plasmid vectors are either vectors currently used for targeted transformation of filamentous fungi e.g. such as pAN7 (Punt et al, 1987 Gene 36:117-124) and other vectors that are well known by the skilled artisan or plasmid vectors according to the invention, preferably plasmid vectors according to the invention.

All of the above-mentioned vectors comprising the PKS marker are hereinbelow termed as "PKS vectors".

A PKS vector is also a vector, which comprises a PKS Marker-expression cassette.

All vectors according to the invention not comprising the PKS marker are hereinbelow termed as "non-PKS vectors".

The present invention furthermore encompasses a method for preparing mutated filamentous fungi, comprising the steps of transferring a non-PKS vector or a PKS vector into a filamentous fungi; and selecting clones of said filamentous fungi, which contain at least one genetic marker introduced by said plasmid vector.

The term filamentous fungi as well as preferred fungi for this purpose are those mentioned and defined above.

In a preferred embodiment, the method for preparing mutated filamentous fungi, comprising the following steps

a) transferring a PKS vector into a filamentous fungi; and

b) selecting successfully transformed filamentous fungi by the absence of color (pigment).

As explained above, the absence of color is based on significant reduction in the amount of polyketide synthetase (or in the polyketide syntethase activity or instability of polyketide synthetase mRNA, which is present in a filamentous fungi). The absence of color can be monitored for example by comparing the transformed fungi with the respective wild-type fungi of the same species.

If a PKS vector is transferred into a filamentous fungi, the disruption of the PKS gene leads to a loss of color (pigment) whereby the degree of transformation can be determined easily. Resulting transformants are white in contrast to the colored wild-type. Thus, the selection according to step b) is done by monitoring the absence of color (pigment) in the filamentous fungi. In a preferred embodiment, the absence of pigment is monitored by optical means.

Alternatively, the absence of color results from the reduction of the polyketide synthethase via antisense techniques. The absence of color hereby means a "reduction of color" or, preferably, loss of color. Absence of color means a reduction in color of at least 20%, preferably between 20 and 40%, more preferably between 40 and 60%, especially preferaby between 60 and 80% and most preferably between 80% and 100%.

In a more preferred embodiment, the PKS vector comprises at least an additional selection marker, preferably the hygromycin resistance gene. In a particular preferred embodiment, the selection of the successfully transformed filamentous fungi comprising a PKS vector can be carried out by hygromycin resistance of success-

fully transformed clones and by the absence of pigment of successfully transformed clones. Most preferably, the PKS vector is a vector according to the invention additionally comprising a PKS marker.

In a further embodiment of the invention, the selection of the succesfully transformed filamentous fungi comprising a non-PKS vector can be carried out by hygromycin resistance of successfully transformed clones.

If a non-PKS vector is used, the vector is linearized by a restriction enzyme cutting in the nucleic acid sequence region of element d). Also nucleic acid sequences exceeding 2000 bp can be used what can be disadvantageous as mentioned above. If a PKS vector is used, the plasmid vector is transferred into a filamentous fungi with the proviso that said vector is linearized by a restriction enzyme in PKS marker nucleic acid sequence. Unlike the non-PKS vectors, the nucleic acid sequence to be expressed recombinantly can also be smaller than 400bp.

In addition to the aforementioned selection methods set forth in step a) to c), homologous recombination can be confirmed by PCR based on oligonucleotides preferably derived from the vector sequence flanking the 5' and 3' regions of the gene to be inserted. Specific examples of these primers are given in the examples.

The plasmid vector may be transferred into the filamentous fungi to be transformed by methods familiar to the skilled worker, preferably via protoplast preparation with driselase or driselase and glucanase as lytic enzyme.

The above-mentioned transformation methods can be also realized in a high throughput screening. Using high throughput screening, many different clones are obtained in parallel so that large numbers of successfully tranformed clones of filamentous fungi can be quickly screened.

The term filamentous fungi as well as preferred fungi for this purpose are those mentioned and defined above.

Due to the convenience of the vector, the above-mentioned KO-plasmid preparation, fungi transformation and screening of the mutants can be at least partially automated so that the whole procedure can also be realized in a high throughput screening. Using high throughput system for example for KO-plasmid preparation and DNA amplification by PCR to screen the recombinant mu-

tants, many different clones are obtained in parallel so that large numbers of transformants can be quickly screened.

Mutagenized filamentous fungi, obtainable according to a method mentioned above, are further encompassed by the present invention.

In an alternative embodiment, the method of transforming filamentous fungi based on the use of polyketide synthetase as marker for transformation comprises the following steps:

a) providing a filamentous fungi characterized by the absence of color (pigment), in which the polyketide synthetase gene is modified in such a way that the polyketide synthetase cannot be functionally expressed;

b) transforming the filamentous fungi of step a) with a "sense expression cassette" or a vector comprising the aforementioned expression cassette;

c) selecting successfully transformed filamentous fungi by the presence of pigment (color).

The nucleic acid sequence as defined in b) i to v. is herein below termed as PKS encoding sequence.

The terms "expression cassette" and "genetic control elements" are explained above.

The "sense-expression cassette" set forth in step b) of the above-mentioned method comprises

a) a promotor sequence in functional linkage with a nucleic acid sequence comprising

i. a nucleic acid sequence shown in SEQ ID NO:3, 4 or 5; or

ii. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:6 by back translation; or

iii. a functional equivalent of the nucleic acid sequence set forth in i) which is encoded by an amino acid sequence shown in SEQ ID NO:6 that has at least an identity of 40% with the SEQ ID NO:6; or

iv. a nucleic acid sequence shown in SEQ ID NO:9 or SEQ ID NO: 11;

v. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:10, SEQ ID NO:12 or SEQ ID NO:13 by back translation;

and optionally

b) further genetic control sequences in functionally linked with a nucleic acid sequence according to a).

The expression cassette or vector comprises preferably a polyketide synthetase encoding nucleic acid sequence as set forth in b) i., ii. or iii..

Preferred phytopathogenic filamentous fungi are those mentioned above. Preferred non-phytopathogenic filamentous fungi are fungi of the group consisting of the genera Aspergillus such as Aspergillus parasiticus, Aspergillus nidulans and Wangiella such as Wangiella dermatidis.

The modification of the polyketide synthetase encoding sequence of the respective fungi can be done either by introduction of at least one mutation in the gene encoding a polyketide synthetase or disruption of the gene encoding a polyketide synthetase.

The term "disruption of the PKS marker" means that the PKS marker sequence is disrupted by introducing DNA comprising stop-codons in the PKS marker sequence e.g. by homologous recombination. The respective methods are well known by the skilled artisan.

The term "mutations" of nucleic acid sequences comprises substitutions, additions, deletions, inversions or insertions of one or more nucleotide residues, which have to bring about termination of translation of the corresponding amino acid sequence of the target protein by the substitution, insertion or deletion of one or more amino acids (e.g. a by frame-shift or introduction of stop codon or amendment of nucleic acid sequence). The respective methods are well known by the skilled artisan.

For example, the mutations are carried out in the flanking regions of exon and intron of a PKS gene. These regions can be determined easily by the skilled artisan. For example, in SEQ ID NO:3 the flanking regions between exon are at bp 1022/1023; bp 1067/1068, bp 1361/1362; bp 1067/1068; bp 1361/1362; bp

1067/1068; bp 1361/1362; bp 1416/1417; bp 2399/2400; bp 2447/2448; bp 2675/2676; bp 2738/2739; bp 5744/5745; bp 5792/5793; and/or bp 7205/7206 (Ende 6. exon bp 7205).

The term "functional analogues" is defined above describe, in the present context nucleic acid sequences which are capable of bringing about the expression, in a filamentous fungi, of a polypeptide with the biological activity of polyketide synthetase and which can be deduced from an amino acid sequence by back translation which has at least an identity of 40%, 41%, 42%, 43%, 44%, 45%, 46%, 47%, 48% or 49%, preferably of 50%, 51%, 52%, 53%, 54%, 55%, 56%, 57%, 58%, 59%, 60%, 61%, 62%, 63%, 64%, 65%, 66%, 67%, 68%, 69%, 70%, 71%, 72%, 73%, 74%, 75%, 76%, 77%, 78% or 79%, more preferably of 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, or 90% most preferred of 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98% or 99% with the SEQ ID NO:6.

As explained above, the plasmid vector may be transferred into the filamentous fungi to be transformed by methods familiar to the skilled worker, preferably via protoplast preparation with driselase or driselase and glucanase as lytic enzyme.

The above-mentioned transformation methods can also be realized in a high throughput screening. Using high throughput screening, many different clones are obtained in parallel so that large numbers of successfully tranformed clones of filamentous fungi can be quickly screened.

The invention is now illustrated by the examples which follow, but is not limited thereto.

Examples

The recombinant methods on which the exemplary embodiments which follow are based are now described briefly:

A: General methods

Cloning methods such as, for example, restriction cleavages, DNA isolation, agarose gel electrophoresis, purification of DNA fragments, transfer of nucleic acids to nitrocellulose and nylon membranes, linking of DNA fragments, transformation of E. coli cells, bacterial cultures, sequence analysis of recombinant DNA and Southern and Western Blots were carried out as described by Sambrook et al., Cold Spring Harbor Laboratory Press (1989) and Ausubel, F.M. et al., Current Protocols in Molecular Biology,

Greene Publishing Assoc. and Wiley-Interscience (1994); ISBN 0-87969-309-6.

The bacterial strains used hereinbelow (E. coli DH5 or XL1 blue) were obtained from Life Technologies or Stratagene . The vectors were used for cloning. DSM:4527 can be used as F. Graminearum wild-type strain 8/1. Restriction maps of the vectors pUCmini-Hyg and PUCmini-Hyg TA are given in Fig 1 and 2.

B: Sequence analysis of recombinant DNA

Recombinant DNA molecules were sequenced using an ABI laser fluorescence DNA sequencer following the method of Sanger (Sanger et al., Proc. Natl. Acad. Sci. USA, 74, 5463-5467(1977)). Fragments resulting from a polymerase chain reaction were sequenced and verified in order to avoid polymerase errors in constructs to be expressed.

C: Materials used

Unless otherwise specified in the text, all of the chemicals used were obtained in analytical grade quality from Fluka (Neu-Ulm), Merck (Darmstadt), Roth (Karlsruhe), Serva (Heidelberg) and Sigma (Deisenhofen). Solutions were prepared using pure pyrogen-free water, referred to in the following text as H_2O, from a Milli-Q water system purification unit (Millipore, Eschborn). Restriction enzymes, DNA-modifying enzymes and molecular-biological kits were obtained from AGS (Heidelberg), Amersham (Brunswick), Biometra (Göttingen), Roche (Mannheim), Genomed (Bad Oeynnhausen), New England Biolabs (Schwalbach/Taunus), Novagen (Madison, Wisconsin, USA), Perkin-Elmer (Weiterstadt), Pharmacia (Freiburg), Qiagen (Hilden) and Stratagene (Heidelberg). Unless otherwise specified, they were used following the manufacturer's instructions.

All of the media and buffers used for the genetic engineering experiments were sterilized either by filter sterilization or by heating in an autoclave.

In degenerated primer sequences, the following abbreviations are used:

A or T = "W";
G or C = "s";
T or C = "Y";
A or C = "M";
A or G = "R";

Examples

Example 1 - Construction of pUCmini-Hyg and PUCmini-Hyg TA vector

A 2536 bp DNA fragment corresponding to the promoter of glycerol-3-phosphate dehydrogenase (GPD1) from *Cochliobolus heterotrophus* associated to the hygromycine B resistance gene from *Escherichia coli* was amplified by PCR with the oligonucleotides

P1 5' atgaagcttggggtttgagggccaatggaacgaaactagtgtaccacttgacc 3' (SEQ ID NO 14); and

P2 5'gacagatctggcgccattcgccattcag 3' (SEQ ID NO 15)

using pGUS5 as template (Mönke, E. and Schäfer, W., 1993, Mol. Gen. Genet. 241: 73-80). The PCR is done using standard protocols; e.g. as described in Maniatis et al., Mol. Cloning.

The resulting DNA fragment was inserted in the plasmid pFDX3809 (WO 01/38504) by the restriction site Hind III and Bgl II introduced by the oligonucleotides P1 and P2. The resulting plasmid pHygB serves as template for a further PCR, wherein the Oligonucleotides

ANK 518 5' ggaatcggtcaatacactac 3' (SEQ ID NO 16)

ANK 519 5' tgtagatctctattcctttgccctcggacgagt 3' (SEQ ID NO 17)

are used to shorten the hygromycin B resistance gene specifically. The resulting PCR fragment comprising 575 bp of the 3'end of the hygromycine gene was inserted in the plasmid pHygB via the restriction sites Nde I/ Bgl II generating the plasmid pHygB-NOS.

A Hind III / Ssp I DNA fragment of 2019 bp containing the expression cassette GPD1 promoter, the hygromycine B resistance gene and the nopaline synthase terminator was isolated from pHygB-NOS and inserted in the pUCmini plasmid (= plasmid pFDX3809, see WO 01/38509) previously treated with EcoRI and HindIII restriction enzymes to give the plasmid pUCmini-Hyg; to do so , the EcoRI ends were made compatible with Ssp I by a fill-in treatment using the Klenow fragment of DNA polymerase I. A second version of pUCmini-Hyg, called pUCmini-Hyg-TA, was obtained by the insertion of the following adaptor in the NotI/AscI restriction sites of pUCmini-Hyg:

5' GGCCGCCACGGATATCTTGGCCAAAGAATTCCTGG 3' (SEQ ID NO 18)

3' CGGTGCCTATAGAACCGGTTTCTTAAGGACCGCGC 5' (SEQ ID NO 19)

The adaptor contains 2 XcmI restriction sites so that XcmI digest of pUCmini-Hyg-TA creates T-overhangs that permits direct cloning of PCR products made with the classical Taq-polymerases.

Example 2 - Construction of the PKS comprising vector "pUCmini-Hyg-PKS"

The nucleic acid sequence encoding PKS was amplified by PCR with degenerated primers

LC1 5'-GAY CCI MGI TTY TTY AAY ATG-3' (SEQ ID NO 20)

LC2c 5'-GTI CCI GTI CCR TGC ATY TC-3' (SEQ ID NO 21)

based on the conserved amino acid sequence of the PKS gene sequences from Aspergillus nidulans, Colletotrichum lagenarium, Penicillium patulum, and Aspergillus parasiticus (Bingle et al., 1999) using genomic DNA of Fusarium graminearum as template. Thermal cycling parameters consisted of an initial denaturation at 94°C for 3 min followed by 34 cycles of 94°C for 1 min (denaturation), 55°C for 1 min (annealing), 72°C for 3 min (extension) and a final extension at 72°C for 10 min according to standard procedures. The resulting PCR product was cloned into the pGEM-T vector (Promega, Mannheim, Germany) to give the plasmid pGEM-T/PKS833 and sequenced. A 633 bp DNA fragment (2236bp to 2870bp of SEQ ID NO:1; corresponding to 2234bp to 2865bp of SEQ ID NO:3; set forth in SEQ ID NO:18) was amplified by PCR using the oligonucleotides

ANK593 5' ATAAGAATGCGGCCGCAATGGCCCTCGAAACAGC 3' (SEQ ID NO 22)

ANK594 5' AAATGGCGCGCCGCGCCCAGAATGACACC 3' (SEQ ID NO 23)

and cloned into the plasmid pUCmini-Hyg using the restriction site NotI and AscI present in the oligonucleotide sequences. The resulting plasmid pUCmini-Hyg-PKS is used for homologous recombination.

The flanking regions of the PKS DNA fragment were obtained by inverse PCR (Triglia T, Peterson MG, Kemp DJ, Nucleic Acids Res 1988 Aug 25;16(16):8186). Genomic DNA was treated with the restriction enzymes PstI, NcoI, or XhoI respectively. DNA was then self-ligated to get circular DNA molecule. The latter was used as template for the inverse PCR reaction using the primers

P1A: 5' TGCCACCTGTAGTCTGCAATCAG 3'(SEQ ID NO 24) and

P2A:5' TGACTAACCCTGACAACTTCGCTG 3' (SEQ ID NO 25)

deduced from the polyketide synthetase (PKS) DNA fragment of the plasmid pGEM-T/PKS833 described above.

In a second step, the PCR product was reamplified with the nested primers

P1B:5' CCAGGATCCGACTGCTCAG 3' (SEQ ID NO 26) and

P2B:5' CTACATCGAGATGCACGGCAC 3' (SEQ ID NO 27)

(deduced from the PKS DNA fragment of the plasmid pGEM-T/PKS833), cloned into the pPCR-XL-TOPO vector (Invitrogen) and sequenced to get SEQ ID NO:1.

Identification of the genomic DNA Sequence

The remaining parts of the flanking regions were obtained by Tail-PCR (Liu YG, Whittier RF; Genomics 1995 Feb 10;25(3):674-81) using 9 arbitrary degenerated primers

FJM-tail-AD1 5'-NGT CGA SWG ANA WGA A-3' (SEQ ID NO 28),

FJM-tail-AD2 5'-GTN CGA SWC ANA WGT T-3' (SEQ ID NO 29),

FJM-tail-AD3 5'-WGT GNA GWA NCA NAG A-3' (SEQ ID NO 30),

FJM-tail-AD4 5'-NTC GAS TWT SGW GTT-3 (SEQ ID NO 31)'

FJM-tail-AD6 5'-TGW GNA GWA NCA SAG A-3' (SEQ ID NO 32),

FJM-tail-AD7 5'-AGW GNA GWA NCA WAG G-3' (SEQ ID NO 33),

FJM-tail-AD8 5'-CAW CGI CNG AIA SGA A-3' (SEQ ID NO 34) and

FJM-tail-AD9 5'-TCS TIC GNA CIT WGG A-3 (SEQ ID NO 35)'

coupled to the primer

TailPKS1c 5'-TTG TTA CTG GAG AGG TAA TGA AG-3' (SEQ ID NO 36)

specific for the 5' PKS flanking region deduced from SEQ ID NO:1,

or coupled to the primer

TailPKS2c 5'-TGA GAC AGA TCT CGC GAG CCC TC-3' (SEQ ID NO 37)

specific for the 3' PKS flanking region deduced from SEQ ID NO:1. After subcloning and subsequent sequencing of the PCR products SEQ ID NO:3 was obtained.

Identification of the cDNA Sequence of Polyketide Synthetase

The PKS cDNA sequence was obtained by RT-PCR with a crude RNA preparation from Fusarium graminearum and various primers deduced from the genomic sequence. This was done according the classicals methods (Ausubel, F.M. et al., Current Protocols in Molecular Biology, Greene Publishing Assoc. and Wiley-Interscience (1994); ISBN 0-87969-309-6). Alignement of cDNA and genomic PKS sequences permits to be identified precisely the location of introns in the genomic sequence.

Example 3 Transformation of F. graminearum

50 ml of CM-medium (Leach et al., 1982, J. Gen. Microbiol. 128: 1719-1729) were inoculated with approximately 10^5 conidia, and incubated for 2 days at 28°C, 140 rpm. Resulting hyphae were homogenized in a Warring-Blender; 200 ml CM were inoculated with 10 ml hyphal suspension, and incubated overnight at 24°C. Mycel were trapped on a sterile filter, and washed two times with sterile water. 2 g of the hyphae were resuspended in 20 ml Driselase/Glucanase (Interspex Products, San Maneo, USA; 5% / 3% in 700 mM NaCl, pH 5.6), and digested 2½ to 3 h at 28°C, 75 rpm. Undigested hyphal were removed from the protoplast suspension by filtration through gauze and Nybold membrane (50 µm pore size). The protoplast suspensions were combined with 700 mM NaCl and again passed through the gauze and the Nybold membrane. The protoplasts were pelleted by centrifugation (1300 x g) in a swing-out rotor and washed two times with ice-cold NaCl 700 mM and centrifuged (830 x g). Then the protoplasts were resuspended in STC (0.8 M sorbitol, 50 mM Tris-HCl pH 8.0, 50 mM $CaCl_2$) and stored on ice until transformation (maximum 1 week).

For transformation, protoplasts were resuspended in 4 parts STC and 1 part SPTC (0.8 M sorbitol, 40% polyethylene glycol 4000, 50 mM Tris-HCl pH 8.0, 50 mM $CaCl_2$) at a concentration of 0.5-2 x 10^8/ml; 30 µg of the pUCmini-Hyg-PKS plasmid DNA linearized with the *Eco*47III restriction site inside the PKS fragment and 5 µl heparin (5 mg/ml in STC) were added to 100 µl of the protoplast suspension in 10 ml tubes. After mixing, samples were incubated

on ice for 30 min. 1 ml SPTC was mixed with the suspension and incubated at room temperature for 20 min. Protoplasts were mixed gently into 200 ml regeneration medium (0.1% (w/v yeast extract, 0.1% (w/v. caseinhydrolysate, 34.2% (w/v) sucrose, 1.6% (w/v) granulated agar) at 43°C and spread on 94 mm plates (20 ml per plate). The plates were incubated at 28°C. After 12-24 h, the plates were overlaid with 10 ml per plate water based selective medium (16g/l granulated agar, 100mg/l hygromycin and further incubated at 28°C until transformants were obtained, which were transferred to fresh CM-Hyg-plates (consisting of CM-media, 100 µg/ml hygromycin and 2% (w/v) agar. The transformants were isolated by single spore isolation. For generation of conidia, the transformants were cultivated on SNA plates (Nirenberg, 1981, Canadian J. Botany 59: 1599-1609) under UV-light 7-14 days at 18°C. Dilutions of conidia were plated on CM-Hyg plates, and single colonies were transferred from these plates to fresh CM-Hyg plates.

Example 4 Southernblot analysis

Genomic DNA was isolated from frozen hyphal material using the Puregene Genomic DNA Isolation Kit (Gentra Systems, Minneapolis USA) and digested for 6 h with *Nru*I restriction enzyme. The genomic DNA was separated by electrophoresis on a 1% (w/v) agarose gel and blotted onto a nylon membrane (Hybond NX; Amersham Pharmacia Biotech, Buckinghamshire, England). A digoxigenin labeled probe was generated by PCR based on specific primers PKS forward 5'-GCG CTT GAG ATG GCT AGT ATC G-3' and and PKS reverse 5'-GTG CCG TGC ATC TCG ATG TAG-3' using pGEM-T/PKS833 as template and digoxigenin labeled dUTPs by PCR reaction according to the recommendation of the manufacturer (Roche Diagnostics GmbH, Mannheim). PCR conditions were 94°C for 3 min (initial denaturation) followed by 30 cycles of 94°C for 30 sec (denaturation), 55°C for 45 sec (annealing), 72°C for 1 min (extension) and a final extension at 72°C for 10 min. The non-radioactive hybridization and the detection were done under highly stringent conditions as described in Roche Molecular Biochemicals DIG Application Manual for Filter Hybridization (Roche Diagnostics GmbH, Mannheim).

To confirm the insertion of the vector construct into the PKS locus in comparison with the wild type gene, primers

EF-PKS 5' atgtctccaaaggaagctgagc 3' (SEQ ID NO 38); and

ER-PKS 5'tcgagtgatggatactgcttcg 3' (SEQ ID NO 39)

are constructed based on the PKS DNA sequence from the plasmid pGEM-T/PKS833; four universal primers are constructed, wherein

Lac 92 5' cggctacactagaaggacagtatttggta 3' (SEQ ID NO 40)

Lac 93 5' gtcaggcaactatggatgaacgaaatagac 3' (SEQ ID NO 41)

Lac 94 5' acccatctcataaataacgtcatgc 3' (SEQ ID NO 42); and

Lac 95 5' caactctatcagagcttggttga 3' (SEQ ID NO 43)

permit amplification of a 412 bp DNA fragment of the hygromycin cassette.

PCR reactions were conducted in classical conditions: 94°C for 3 min (initial denaturation) followed by 30 cycles of 94°C 60 sec (denaturation), 55°C for 90 sec (annealing), 72°C for 90 sec (extension) and a final extension at 72°C for 10 min.
Six (6) recombinant clones resistant to hygromycine were analyzed by PCR using the primer set Lac 94 / Lac 95 specific for the hygromycin resistance gene. All the mutants were found to present the expected DNA fragment of 412 bp, indicating the integration of the plasmid pUCmini-Hyg-PKS in the genome.

A 712 bp corresponding to the PKS gene could be amplified with the primer set EF-PKS/ER-PKS mentioned above using genomic DNA from a wild type strain; however no PCR fragments were amplified with genomic DNA from the recombinant clones indicating that the PKS gene is disrupted by the insertion of pUCmini-Hyg-PKS. This was confirmed by PCR amplification EF-PKS combined with Lac 93 (hybridizing to the plasmid backbone near Not I restriction site) and ER-PKS combined with Lac 92 (hybridizing to the plasmid backbone near Asc I restriction site). In both cases, DNA fragments of about 600 bp were amplified for the recombinant clones but not for the wild type strain (WT). Alltogether, the PCR analysis using the different primer sets proves that the plasmid pUCmini-Hyg-PKS was targeted specifically in the PKS locus by homologous recombination. This process permits the PKS gene to be disrupted since the recombinant mutants were found to lack the typical pigmentation (purple) of the wildtype strain.

Example 5 functional expression of Green Fluorescent Protein (GFP) in Fusarium graminearum

A) Plasmid construction

In a first step, a 67bp DNA fragment encoding the peptide leader of the first 23 amino acids from N-terminus of the yeast ARH1 (SwissProt;P48360) was amplified by PCR using the primers

Lac 80 5' cccgaattcatgagctttgttcaaataagg 3' (SEQ ID NO 44) and

Lac 81 5' ttattctagattttccatgggaatggatacagtcttacg 3' (SEQ ID NO 45)

In a second step, a 734 bp DNA fragment encoding the Green Fluorescent Protein (GFP) was amplified by PCR using the plasmid pEGFP-N2 (Genbank; U57608) and the primers

Lac 84 5' cgccaccatggtgagcaagggcgaggagctgtt 3' (SEQ ID NO 46) and

Lac 85 5' tatgatctagagtcgcggccgctttacttgtacagctcg 3' (SEQ ID NO 47).

The PCR products were assembled in frame with the Nco I restriction sites present in the oligonucleotides Lac 81 and Lac 84 and cloned in the expression plasmid pYes2 (Invitrogen) using the restriction sites EcoRI and Xba I present in the oligonucleotides Lac 80 and Lac 85, respectively. In the resulting plasmid pLAC7, the recombinant gene encoding GFP is under the control of the galactose (Gal 1) promoter and cytochrome C1 terminator.

A 2892 bp DNA fragment containing the GFP expression cassette was isolated from pLac7 using the restriction sites Nae I and Bsa I and cloned in the plasmid pUCmini-Hyg-PKS (see example 2). To do so , pUCmini-Hyg-PKS was firstly cut by Asc I and filled in according to classical method then treated with Bsa I. The resulting plasmid pUCmini-Hyg-PKS-GFP contains all genetic elements permitting the production of recombinant GFP in Fusarium graminearum.

B) Transformation of Fusarium graminearum with pUCmini-Hyg-PKS-GFP and analysis of transformants

The transformation was done as described in example 3, wherein pUCmini-Hyg-PKS-GFP was linearized with EcoR47III. The correct integration of the plasmid in the PKS locus was observed after single conidiation by the absence of pigmentation of the recombinant mutants.

In addition, the integration was confirmed by PCR as described in example 4 using the following primer combinations EF-PKS (see example 4; SEQ ID NO:38) and ER-PKS (see example 4; SEQ ID NO:39), whereby no amplification were observed since the gene PKS is disrupted, whereas wild type strain or unspecific mutants presented a 714 bp DNA fragment corresponding to the expected PKS DNA fragment.

Using the primer combination EF-PKS (see example 4; SEQ ID NO:38) and

Lac 211 5' gcttctaatccgtactagtggatca 3' (SEQ ID NO 48)

the amplification of a 835 bp DNA corresponding to the 5' end plasmid integration in the PKS locus of the mutants was observed. No DNA fragment was amplified for the wildtype strain due the absence of the DNA sequence complementary to Lac211.

The primer combination

ANK 458 5' ctttgatcttttctacggggtctga 3' (SEQ ID NO 49) and

ER-PKS (see example 4; SEQ ID NO:39) led to the amplification of a 718 bp DNA corresponding to the 3' end plasmid integration in the PKS locus of the mutants. No DNA fragment was amplified for the wildtype strain due the absence of the DNA sequence complementary to ANK 458.

C) Detection of the production of GFP

The recombinant mutants were grown for a few days in CM-Hyg medium as described in example 3 except for glucose which was replaced by galactose as a carbon source. The fluorescence of GFP was detected using the polarstar spectrophotometer (Firma BMG; Ex: 385nm and Em: 520nm). In these conditions fluorescence was observed for the strains which showed integration of the plasmid whereas no fluorescence was observed for the wildtype strains.

Brief description of the figures

Figure 1: Map of pUCmini-Hyg

Figure 1: Map of PUCmini-Hyg TA

Claims

1. A plasmid vector for targeted transformation of filamentous fungi comprising

a) an origin of replication for a host organism not originating from the filamentous fungi to be transformed;

b) a selection marker for a host organism not originating from the filamentous fungi;

c) a promotor facilitating recombinant expression in fungi that is functionally linked to the coding region of the hygromycin resistance gene which is functionally linked to a terminator which facilitates transcription termination in filamentous fungi;

wherein the overall size of the elements a), b) and c) does not exceed 4500 bp; and

d) a nucleic acid sequence which is homologous to nucleic acid sequences of the filamentous fungi to be transformed and makes homologous recombination in the filamentous fungi to be transformed possible.

2. A plasmid vector as claimed in claim 1, wherein the origin of replication a) originates from bacteria.

3. A plasmid vector as claimed in claims 1 to 2, wherein the selection marker b) imparts a resistance to antibiotics.

4. A plasmid vector according to claims 1 to 3, wherein the promotor of element c) is selected from the group consisting of the GPD-1-, PX6-, TEF-, CUP1-, PGK-, GAP1-, TPI, PHO5-, AOX1, GAL10/CYC-1, CYC1, OliC-, ADH-, TDH-, Kex2-, MFa- and the NMT-promotor.

5. A plasmid vector according to claims 1 to 4, wherein the terminator of element c) is selected from the group consisting of the AOX1-, nos-, PGK-, TrpC- and the CYC1-terminator.

6. A plasmid vector according to claims 1 to 5, wherein the promotor of element c) is the GPD-1-promotor and the terminator of element c) is the nos-terminator.

7. A plasmid vector according to claims 1 to 6, wherein the nucleic acid sequence d) is functionally linked to a promotor facilitating recombinant expression in filamentous fungi.

8. A plasmid vector according to claims 1 to 7, wherein the nucleic acid sequence d) is functionally linked to a transcription terminator facilitating recombinant expression in filamentous fungi.

9. A selection marker comprising a nucleic acid sequence encoding a polyketide synthetase fragment, wherein said nucleic acid sequence comprises

i. a nucleic acid sequence shown in SEQ ID NO:1 or SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4 or SEQ ID NO:5; or

ii. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:6 by back translation; or

iii. a functional equivalent of the nucleic acid sequence set forth in i) which is encoded by an amino acid sequence that has at least an identity of 50% with the SEQ ID NO: or from a functional equivalent of an amino acid sequence shown in SEQ ID NO:6 that has at least an identity of 41% with the SEQ ID NO:6 or from a functional equivalent of an amino acid sequence shown in SEQ ID NO:8 that has at least an identity of 49% with the SEQ ID NO:8 or from a functional equivalent of an amino acid sequence shown in SEQ ID NO:10 that has at least an identity of 6; or

iv. parts of the nucleic acid sequence as defined in i., ii. or iii. consisting of at least 300bp; or

v. parts of the nucleic acid sequence as defined in i., ii. or iii. consisting of at least 300bp comprising

a) a nucleic acid sequence shown in SEQ ID NO:7 ; or

b) a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:8 by back translation; or

c) a functional equivalent of a nucleic acid sequence set forth in a), which is encoded by amino acid sequence that has at least an identity of 85% with the SEQ ID NO:8.

10. Use of a nucleic acid sequence comprising

a) a nucleic acid sequence encoding a polyketide synthetase; or

b) parts of the nucleic acid sequence as defined in i. consisting of at least 300bp.

as marker for targeted transformation in filamentous fungi.

11. Use of a nucleic acid sequence according to claim 10 said nucleic acid sequence comprising

i. a nucleic acid sequence according to claim 9; or

ii. a nucleic acid sequence shown in SEQ ID NO:9 or SEQ ID NO: 11; or

iii. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:10, SEQ ID NO:12 or SEQ ID NO:13 by back translation; or

iv. a functional equivalent of the nucleic acid sequence set forth in i), which is encoded by an amino acid sequence that has at least an identity of 40% with the SEQ ID NO:6 or from a functional equivalent of an amino acid sequence shown in SEQ ID NO:6 that has at least an identity of 38% with the SEQ ID NO:6 or from a functional equivalent of an amino acid sequence shown in SEQ ID NO:8 that has at least an identity of 39% with the SEQ ID NO:8 or from a functional equivalent of an amino acid sequence shown in SEQ ID NO:10 that has at least an identity of ; or

v. parts of the nucleic acid sequence as defined in ii., iii. or iv. consisting of at least 300bp; or

vi. parts of the nucleic acid sequence as definied in ii., iii or iv. consisting of at least 300bp comprising a nucleic acid sequence, which is encoded by an amino acid

sequence that has at least an identity of 68% with the SEQ ID NO:8.

12. A plasmid vector for targeted transformation of filamentous fungi additionally comprising a selection marker comprising a nucleic acid sequence encoding a polyketide synthetase fragment, said nucleic acid sequence comprising

i. a nucleic acid sequence according to claim 9; or

ii. a functional equivalent of the nucleic acid sequence set forth in i) which is encoded by an amino acid sequence that has at least an identity of 40% with the SEQ ID NO:6.

iii. a nucleic acid sequence shown in SEQ ID NO:9 or SEQ ID NO: 11;

iv. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:10, SEQ ID NO:12 or SEQ ID NO:13 by back translation; or

v. parts of the nucleic acid sequence as defined in ii., iii. or iv. consisting of at least 300bp; or

vi. parts of the nucleic acid sequence as defined in i., ii. or iii. or iv. consisting of at least 300bp, which are encoded by an amino acid sequence that has at least an identity of 68% with SEQ ID NO:8.

13. A plasmid vector for targeted transformation of filamentous fungi as claimed in claims 1 to 8, additionally comprising a selection marker comprising a nucleic acid sequence encoding a polyketide synthetase fragment, said nucleic acid sequence comprising

i. a nucleic acid sequence according to claim 9; or

ii. a functional equivalent of the nucleic acid sequence set forth in i), which is encoded by an amino acid sequence that has at least an identity of 40% with the SEQ ID NO:6; or

iii. a nucleic acid sequence shown in SEQ ID NO:9 or SEQ ID NO: 11;

iv. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:10, SEQ ID NO:12 or SEQ ID NO:13 by back translation; or

v. parts of the nucleic acid sequence as defined in ii., iii. or iv. consisting of at least 300bp; or

vi. parts of the nucleic acid sequence as defined in i., ii. or iii. or iv. consisting of at least 300bp comprising a nucleic acid sequence, which is encoded by a functional equivalent of an amino acid sequence that has at least an identity of 68% with the SEQ ID NO:8.

14. An expression cassette comprising

a) a promotor sequence in functional linkage with a nucleic acid sequence according to claim 9 in antisense orientation; and optionally

b) further genetic control sequences functionally linked to a nucleic acid sequence according to a).

15. A plasmid vector for targeted transformation of filamentous fungi additionally comprising an expression cassette according to claim 14.

16. A plasmid vector for targeted transformation of filamentous fungi as claimed in claims 1 to 8, additionally comprising an expression cassette according to claim 14.

17. A method for transforming filamentous fungi, comprising the following steps

a) transferring a plasmid vector according to claim 12, 13, 15 or 16 into a filamentous fungi;

b) selecting successfully transformed filamentous fungi by the absence of color.

18. An expression cassette comprising

a) a promotor sequence in functional linkage with a nucleic acid sequence comprising

i. a nucleic acid sequence shown in SEQ ID NO:3, 4 or 5; or

ii. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:6 by back translation; or

iii. a functional equivalent of the nucleic acid sequence set forth in i) which is encoded by an amino acid sequence shown in SEQ ID NO:6 that has at least an identity of 40% with the SEQ ID NO:6; or

iv. a nucleic acid sequence shown in SEQ ID NO:9 or SEQ ID NO: 11;

v. a nucleic acid sequence which, owing to the degeneracy of the genetic code, can be deduced from the amino acid sequence shown in SEQ ID NO:10, SEQ ID NO:12 or SEQ ID NO:13 by back translation;

and optionally

b) further genetic control sequences functionally linked to a nucleic acid sequence according to a).

19. A method for transformation of filamentous fungi, comprising the following steps

a) providing a filamentous fungi, in which the polyketide synthetase gene is modified in such away that the polyketide synthetase cannot be functionally expressed;

b) transforming the filamentous fungi of step a) with an expression cassette according to claim 18 or a vector comprising the aforementioned expression cassette;

c) selecting successfully transformed filamentous fungi by the presence of color.

20. A method as claimed in claim 17 or 19, wherein the plasmid vector comprises at least an additional selection marker.

21. A method as claimed in claims 17, 19 or 20, wherein the selection is confirmed by PCR.

22. A method as claimed in claims 17, 19, 20 or 21, wherein the filamentous fungi are succesuflly transformed and identified in a high-throughput screening.

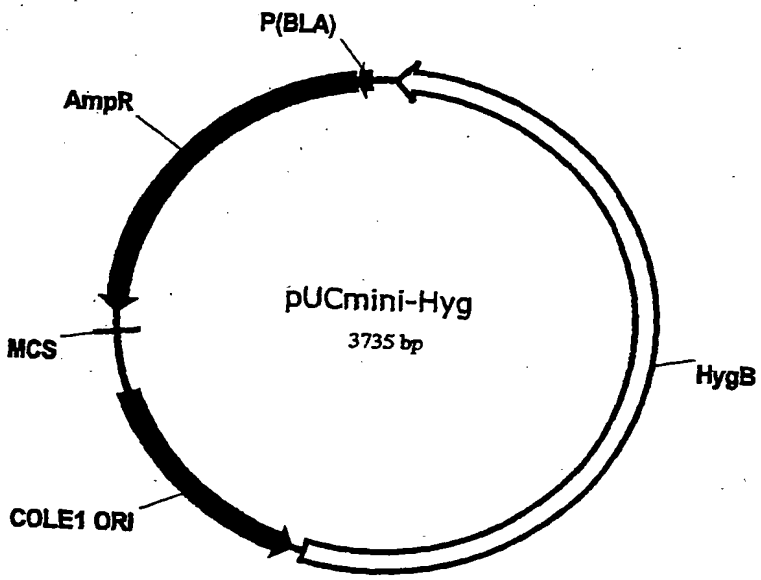

Figure 1

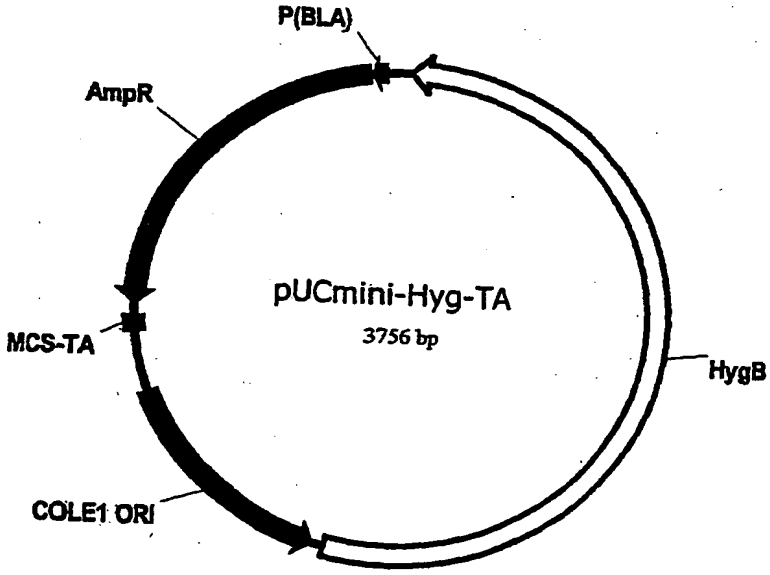

Figure 2

SEQUENCE LISTING

<110> BASF Aktiengesellschaft

<120> Plasmid vectors for transformation of filamentous fungi

<130> Polyketide Synthetase

<140>
<141>

<160> 49

<170> PatentIn Ver. 2.1

<210> 1
<211> 5888
<212> DNA
<213> Fusarium graminearum

<400> 1
atcgagtttc gtgttgcgtg tcatggtgta gcgaacaata ccatactcct ccatcaaagg 60
gctgacgagc ttggcatgga cctttgtcat atggtgcctg tactctttct cgctaagaga 120
aggtttttg taggccgtga tggttaggca aacaagggta tcaggttgac cggtgacagt 180
tgcctcatga gaggtttctt gggtatcact atgaggcgac atgatgcagg ttggagtttg 240
ttatcgaagt gctgggtagg atttggttga cagagttgca taaactggag ctctggatac 300
gcaaggggtc ataagagtat ggatacgccg tcgattagaa gctgtcagat ccagtgcatc 360
aatggcccga atgacgagag attgctgctc ggcactgaaa gtctgcagcg tgcttgtttc 420
ggagttatct ctacagctgc accgaatgac gaacaagccc gactatccat gtccgacgag 480
cccggaccga cgtgatccga atgctgtcta acacttacca agacaatcat ggggtatctc 540
tgaattatga ttgtgttgct caagcagcta atcccaactc ttaatcacgt caagtgccat 600
gcttacctgt attatcatct gtcatcaaca taccaactcc tatatatagg ggattgtgct 660
tcattacctc tccagtaaca actacttgga gttcaaacca catttcaact tactctttta 720
gttcattcaa catgacccca tcaatgatgg aggtattcgt ttttggggac caaagcacac 780
gctttgcccc tccactgaaa gacctactcc tcaaaggcaa cagtccttac ttgacacatt 840
ttgttaaaca agttcacgca cttcttagaa gggagatatc atccttgccg gcagttcaac 900
agaagctttt cccaaacttt gccgacattc aggaactcgt ctccaagtca gattggggca 960
gtggtaaccc tgctttgaca agcgctttag catgctttta ccatctttgc agtttcattc 1020
agtgagtaac ataaccaagt acccatagat ctctcctgac catccagcct tttacgatgg 1080
acaaggtcgt acctttcctt cggagaacag tcgcattatt ggactttgcg ttggttcact 1140
cgctgctact gctgtcagtt gctccacatc actgagtgaa ttggtatcag ctggtgtaga 1200
tgctgttcgt gtggcattgc acgtcggact acgggtatgg cgaactacct ccctttcga 1260
tgtaccagac aggccctccg ccacttggtt cataattgtg cccgaggcag tactaccaag 1320
agaatctgcg caagaccgac ttgactcatt catcattgaa atggtaagca attctttgtc 1380
cttggagctc tcataaaaca tattcactct tctttaggga cttgctcgat catcagttcc 1440
ttacatcagc tcggtcgcac atcacaacat gaccatcagt ggtccaccat ccgtcctcga 1500
aaagttcatt cacagtatat caacatcacc gaaagattct cttccagtgc cgatctatgc 1560
tccgtaccac gccagccatc tttacagcat ggatgatgta gacgaggtcc ttagcctgtc 1620
tgcaccttct tttgcatcag agtccatcat tccactcatt tcaagctcct cgggtgacga 1680
gttacagcca ctcaagtatg cagatctact ccgctgctgt gttagtgata tgctcataca 1740
gccactggat cttaccaagg tctcacaagc agtggcccag cttctcgagg ttagctcatc 1800
tacacgtgcc ataataaagc ctatagcaac cagcgtctcc aacagtctag tgtctgtttt 1860
ggagccgacg ctagcagaac gatgcgccgt ggacaacagc atggggccca aagcctcgac 1920

cagccactca tcagcagaga cacaaaccga gtcatcaagc aagaactcca aaattgcgat 1980
tgttgctatg tctggtcgct ttccagacgc agctgacttg agtgaattct gggatcttct 2040
ctacgaaggt cgcgatgttc atcgacaaat tcccgaggac cgattcaacg cagagctcca 2100
ttacgacgct actgggcgac gtaagaacac cagcaaggtc atgaatggct gcttcatcaa 2160
ggaaccagga ctgttcgacg ctaggttctt caacatgtct ccaaaggaag ctgagcagtc 2220
ggatcctggc cagcgaatgg ccctcgaaac agcttacgag gcgcttgaga tggctagtat 2280
cgtaccagac agaacacctt cgacacagag agatcgtgtt ggtgtgttct acggcatgac 2340
tagcgatgat tggagagagg tcaacagtgg gcagaatgtc gacacttatt ttattcctgg 2400
taagttgttt gttcgcgttt cacgacaata ctgattgcag actacaggtg gcaacagagc 2460
gttcactcct ggtcgactca actacttctt caagttcagt gggcctagcg ctagtgttga 2520
tacggcttgc tcctccagtc tcgttggctt gcacttggct tgtaattccc tctggagaaa 2580
tgattgcgat acagctattg cgggcggaac caatgtcatg actaaccctg acaacttcgc 2640
tggtttggac cgaggccact tcctatctca agaaccggta agtctctcac agatcttgga 2700
agcttccttc tttttagtgt ttttactgac cttgttccag gcaactgcaa caccttttgac 2760
gatggagcag acggatactg tcgagctgat ggcgtcggaa ccatcatcct caagcggctt 2820
gaggacgccg aagctgacaa tgaccctatt ctcggtgtca ttctgggcgc ttacacaaac 2880
cactcagccg aagcagtatc catcactcga ccacatgccg gagctcaaga gtacatcttc 2940
tccaaactcc tccgtgagtc gggcaccgat ccctacaacg ttagctacat cgagatgcac 3000
ggcacaggca ctcaagccgg cgacgcaacc gagatgacat ccgtcctcaa gacgtttgct 3060
cctaccagcg gcttcggcgg tcgattgcct caccaaaacc ttcacttggg ttcagtcaag 3120
gccaatgtcg ggcacggtga atccgcatct ggtatcattg ctctgatcaa gacgctgctt 3180
atgatggaga agaacatgat cccgccgcat tgtggtatca agacaaagat caatcaccat 3240
tttcctacgg atctcactca gcgcaatgtc catatcgcca aagttccgac atcttggaca 3300
agatcgggtc aagccaatcc acgcattgct ttcgtcaata acttctctgc cgctggtggt 3360
aactctgctg tcctactgca agatgctcct cagccatcgg tagtttcgga tgttacagac 3420
cctcgcacat cccatgttgt cactatgtcc gctcgatcag cagattccct caggaagaac 3480
ctcgccaatc tcaaggagct tgtagaaggc caaggtgact cggaggtcgg cttcctgagc 3540
aagctgtcct acacaaccac cgccaggcgc atgcatcatc aattccgagc ttcggtcaca 3600
gcacagactc gtgaacagct gctgaagggc cttgattccg ccattgaacg ccaggatgtg 3660
aagaggatcc ccgccgccgc gccctctgtc ggctttgtgt ttagcggcca aggcgcccaa 3720
taccgtggta tgggcaagga gtactttaca tctttcacag ccttccgctc tgagatcatg 3780
tcttacgaca gtatcgccca agcccaaggc ttcccgtcaa tcctcccact gatccgagga 3840
gaggtggaag ctgactcgtt gagtcctgtt gagatccagc tgggtctcac ttgcctgcag 3900
atggcactgg ccaagctatg gaagtcattc ggtgttgagc caggctttgt tctcggacac 3960
agcttaggcc actatgctgc tttacacgtc gctggtgttc tgtccgccaa tgataccatt 4020
tacctcactg gcatcagagc gcagctgctc gtggataagt gccaggcagg aacccactca 4080
atgctggcag tgagggcatc cttactacag atccaacagt tcctcgatgc caacattcac 4140
gaggttgcat gtgtcaatgg ttcacgagaa gtcgtcatca gtggacgcgt tgccgacatt 4200
gaccagctgg ttggcctttt gtcggctgac aacatcaagg cgacccgcgt caaggtgcca 4260
tttgccttcc actcagcgca ggttgacccc attctctccg acttggatac agcggcgtcg 4320
cgcgtcacct tccactccct ccagattcct gttctttgtg cccttgacag ctctgtcatc 4380
agccctggaa accatggtgt cattggtccc cttcatctac agcgacattg tcgtgagaca 4440
gtcaactttg agggtgctct acatgctgcg gagcacgaga agatcatcaa caagacatca 4500
actctatgga tcgagattgg tccccatgtt gtctgctcta ccttcctcaa gtccagcctt 4560
ggtccaagca cacctgctat cgcatcgctt cgccgaaatg acgattgctg gaaggtgttg 4620
gctgatggtt tgagcagtct ctacagcagc gggttgacaa ttgactagaa cgagtatcat 4680
cgcgacttca aggcctctca ccaggtactt cgtctgcctt gttacagctg ggagcacaag 4740
aattactgga tacagtacaa gtacgattgg tccttggcta aaggtgatcc tccaattgcc 4800
cctaacagct cggttgaagc agtctcagct ttatcaacac cctcggtcca gaagattcta 4860

caggagactt cccttgatca ggtattgact atcgttgctg agacagatct cgcgagccct 4920
ctattgtcag aggttgccca aggtcatcgg gtcaacggtg tcaaagtctg cacatcttcc 4980
gtgtacgctg atgttggctt gacgctgggt aagtacattt tggacaacta ccgcaccgac 5040
ttagagggtt atgcggtcga tgttcacggt attgaggtcc acaagccact tcttctcaaa 5100
gaagacatga acggaacgcc ccaggctaca ccgttccgca tcgaagtgcg atacccaatc 5160
cagagcacca cggcgctgat gagcatctcc accactggcc ccaacggtca gcacatcaag 5220
catgctaact gcgaacttcg actcgagcat ccgtcgcaat gggaagcgga gtgggatcgc 5280
caagcctacc tcatcaatcg cagcgtcaac tgccttctgc agcgatcagc acaaggtttg 5340
gacagcatgt tggcaaccgg aatggtctac aaggtcttct cctccctcgt cgactatgcc 5400
gatggctaca agggtctgca ggaggttgtc ttgcacagcc aagagctcga gggcacagca 5460
aaagtgcgct tccaaactcc ctcgggaggt ttcgtctgca atcccatgtg gattgacagc 5520
tgtggtcaga cgaccggctt catgatgaac tgtcatcaga ctacgcccaa tgactacgtc 5580
tacgtcaatc atggctggaa gtcgatgaga ttggccaagg cgttccgtga agatggtacc 5640
tatcggactt atatccggat gaggcccatt gatagcacca agttcgctgg tgacttgtac 5700
attcttgatg aggatgacac tgtggttggt gtttatggag acataacaat gagtaacacg 5760
aatctttgtc aacaacaatg ctaacgtaaa gaacagttcc aaggtttgcc gcgacgagtt 5820
ctcaacacag tcctgccatc tgccaacgcg gttccagttg atgctcccat ggttcaagac 5880
gacctcca 5888

<210> 2
<211> 5157
<212> DNA
<213> Fusarium graminearum

<400> 2
atgaccccat caatgatgga ggtattcgtt tttggggacc aaagcacacg ctttgcccct 60
ccactgaaag acctactcct caaaggcaac agtccttact tgacacattt tgttaaacaa 120
gttcacgcac ttcttagaag ggagatatca tccttgccgg cagttcaaca gaagcttttc 180
ccaaactttg ccgacattca ggaactcgtc tccaagtcag attggggcag tggtaaccct 240
gctttgacaa gcgctttagc atgcttttac catctttgca gtttcattca gtgagtaaca 300
taaccaagta cccatagatc tctcctgacc atccagcctt ttacgatgga caaggtcgta 360
cctttccttc ggagaacagt cgcattattg gactttgcgt tggttcactc gctgctactg 420
ctgtcagttg ctccacatca ctgagtgaat tggtatcagc tggtgtagat gctgttcgtg 480
tggcattgca cgtcggacta cgggtatggc gaactacctc ccttttcgat gtaccagaca 540
ggccctccgc cacttggttc ataattgtgc ccgaggcagt actaccaaga gaatctgcgc 600
aagaccgact tgactcattc atcattgaaa tggtaagcaa ttctttgtcc ttggagctct 660
cataaaacat attcactctt ctttagggac ttgctcgatc atcagttcct tacatcagct 720
cggtcgcaca tcacaacatg accatcagtg gtccaccatc cgtcctcgaa aagttcattc 780
acagtatatc aacatcaccg aaagattctc ttccagtgcc gatctatgct ccgtaccacg 840
ccagccatct ttacagcatg gatgatgtag acgaggtcct tagcctgtct gcaccttctt 900
ttgcatcaga gtccatcatt ccactcattt caagctcctc gggtgacgag ttacagccac 960
tcaagtatgc agatctactc cgctgctgtg ttagtgatat gctcatacag ccactggatc 1020
ttaccaaggt ctcacaagca gtggcccagc ttctcgaggt tagctcatct acacgtgcca 1080
taataaagcc tatagcaacc agcgtctcca acagtctagt gtctgttttg gagccgacgc 1140
tagcagaacg atgcgccgtg gacaacagca tggggcccaa agcctcgacc agccactcat 1200
cagcagagac acaaaccgag tcatcaagca agaactccaa aattgcgatt gttgctatgt 1260
ctggtcgctt tccagacgca gctgacttga gtgaattctg ggatcttctc tacgaaggtc 1320
gcgatgttca tcgacaaatt cccgaggacc gattcaacgc agagctccat tacgacgcta 1380
ctgggcgacg taagaacacc agcaaggtca tgaatggctg cttcatcaag gaaccaggac 1440
tgttcgacgc taggttcttc aacatgtctc caaaggaagc tgagcagtcg gatcctggcc 1500
agcgaatggc cctcgaaaca gcttacgagg cgcttgagat ggctagtatc gtaccagaca 1560

gaacaccttc gacacagaga gatcgtgttg gtgtgttcta cggcatgact agcgatgatt 1620
ggagagaggt caacagtggg cagaatgtcg acacttattt tattcctggt aagttgtttg 1680
ttcgcgtttc acgacaatac tgattgcaga ctacaggtgg caacagagcg ttcactcctg 1740
gtcgactcaa ctacttcttc aagttcagtg ggcctagcgc tagtgttgat acggcttgct 1800
cctccagtct cgttggcttg cacttggctt gtaattccct ctggagaaat gattgcgata 1860
cagctattgc gggcggaacc aatgtcatga ctaaccctga caacttcgct ggtttggacc 1920
gaggccactt cctatctcaa gaaccggtaa gtctctcaca gatcttggaa gcttccttct 1980
ttttagtgtt tttactgacc ttgttccagg caactgcaac acctttgacg atggagcaga 2040
cggatactgt cgagctgatg gcgtcggaac catcatcctc aagcggcttg aggacgccga 2100
agctgacaat gaccctattc tcggtgtcat tctgggcgct tacacaaacc actcagccga 2160
agcagtatcc atcactcgac cacatgccgg agctcaagag tacatcttct ccaaactcct 2220
ccgtgagtcg ggcaccgatc cctacaacgt tagctacatc gagatgcacg gcacaggcac 2280
tcaagccggc gacgcaaccg agatgacatc cgtcctcaag acgtttgctc ctaccagcgg 2340
cttcggcggt cgattgcctc accaaaacct tcacttgggt tcagtcaagg ccaatgtcgg 2400
gcacggtgaa tccgcatctg gtatcattgc tctgatcaag acgctgctta tgatggagaa 2460
gaacatgatc ccgccgcatt gtggtatcaa gacaaagatc aatcaccatt ttcctacgga 2520
tctcactcag cgcaatgtcc atatcgccaa agttccgaca tcttggacaa gatcgggtca 2580
agccaatcca cgcattgctt tcgtcaataa cttctctgcc gctggtggta actctgctgt 2640
cctactgcaa gatgctcctc agccatcggt agtttcggat gttacagacc ctcgcacatc 2700
ccatgttgtc actatgtccg ctcgatcagc agattccctc aggaagaacc tcgccaatct 2760
caaggagctt gtagaaggcc aaggtgactc ggaggtcggc ttcctgagca agctgtccta 2820
cacaaccacc gccaggcgca tgcatcatca attccgagct tcggtcacag cacagactcg 2880
tgaacagctg ctgaagggcc ttgattccgc cattgaacgc caggatgtga agaggatccc 2940
cgccgccgcg ccctctgtcg gctttgtgtt tagcggccaa ggcgcccaat accgtggtat 3000
gggcaaggag tactttacat ctttcacagc cttccgctct gagatcatgt cttacgacag 3060
tatcgcccaa gcccaaggct tcccgtcaat cctcccactg atccgaggag aggtggaagc 3120
tgactcgttg agtcctgttg agatccagct gggtctcact tgcctgcaga tggcactggc 3180
caagctatgg aagtcattcg gtgttgagcc aggctttgtt ctcggacaca gcttaggcca 3240
ctatgctgct ttacacgtcg ctggtgttct gtccgccaat gataccattt acctcactgg 3300
catcagagcg cagctgctcg tggataagtg ccaggcagga acccactcaa tgctggcagt 3360
gagggcatcc ttactacaga tccaacagtt cctcgatgcc aacattcacg aggttgcatg 3420
tgtcaatggt tcacgagaag tcgtcatcag tggacgcgtt gccgacattg accagctggt 3480
tggccttttg tcggctgaca acatcaaggc gacccgcgtc aaggtgccat ttgccttcca 3540
ctcagcgcag gttgacccca ttctctccga cttggataca gcggcgtcgc gcgtcacctt 3600
ccactccctc cagattcctg ttctttgtgc ccttgacagc tctgtcatca gccctggaaa 3660
ccatggtgtc attggtcccc ttcatctaca gcgacattgt cgtgagacag tcaactttga 3720
gggtgctcta catgctgcgg agcacgagaa gatcatcaac aagacatcaa ctctatggat 3780
cgagattggt ccccatgttg tctgctctac cttcctcaag tccagccttg gtccaagcac 3840
acctgctatc gcatcgcttc gccgaaatga cgattgctgg aaggtgttgg ctgatggttt 3900
gagcagtctc tacagcagcg ggttgacaat tgactagaac gagtatcatc gcgacttcaa 3960
ggcctctcac caggtacttc gtctgccttg ttacagctgg gagcacaaga attactggat 4020
acagtacaag tacgattggt ccttggctaa aggtgatcct ccaattgccc ctaacagctc 4080
ggttgaagca gtctcagctt tatcaacacc ctcggtccag aagattctac aggagacttc 4140
ccttgatcag gtattgacta tcgttgctga gacagatctc gcgagccctc tattgtcaga 4200
ggttgcccaa ggtcatcggg tcaacggtgt caaagtctgc acatcttccg tgtacgctga 4260
tgttggcttg acgctgggta agtacatttt ggacaactac cgcaccgact tagagggtta 4320
tgcggtcgat gttcacggta ttgaggtcca caagccactt cttctcaaag aagacatgaa 4380
cggaacgccc caggctacac cgttccgcat cgaagtgcga tacccaatcc agagcaccac 4440
ggcgctgatg agcatctcca ccactggccc caacggtcag cacatcaagc atgctaactg 4500

cgaacttcga ctcgagcatc cgtcgcaatg ggaagcggag tgggatcgcc aagcctacct 4560
catcaatcgc agcgtcaact gccttctgca gcgatcagca caaggtttgg acagcatgtt 4620
ggcaaccgga atggtctaca aggtcttctc ctccctcgtc gactatgccg atggctacaa 4680
gggtctgcag gaggttgtct tgcacagcca agagctcgag ggcacagcaa aagtgcgctt 4740
ccaaactccc tcgggaggtt tcgtctgcaa tcccatgtgg attgacagct gtggtcagac 4800
gaccggcttc atgatgaact gtcatcagac tacgcccaat gactacgtct acgtcaatca 4860
tggctggaag tcgatgagat tggccaaggc gttccgtgaa gatggtacct atcggactta 4920
tatccggatg aggcccattg atagcaccaa gttcgctggt gacttgtaca ttcttgatga 4980
ggatgacact gtggttggtg tttatggaga cataacaatg agtaacacga atctttgtca 5040
acaacaatgc taacgtaaag aacagttcca aggtttgccg cgacgagttc tcaacacagt 5100
cctgccatct gccaacgcgg ttccagttga tgctcccatg gttcaagacg acctcca 5157

<210> 3
<211> 7339
<212> DNA
<213> Fusarium graminearum

<400> 3
atcgagtttc gtgttgcgtg tcatggtgta gcgaacaata ccatactcct ccatcaaagg 60
gctgacgagc ttggcatgga cctttgtcat atggtgcctg tactctttct cgctaagaga 120
aggttttttg taggccgtga tggttaggca aacaagggta tcaggttgac cggtgacagt 180
tgcctcatga gaggtttctt gggtatcact atgaggcgac atgatgcagg ttggagtttg 240
ttatcgaagt gctgggtagg atttggttga cagagttgca taaactggag ctctggatac 300
gcaaggggtc ataagagtat ggatacgccg tcgattagaa gctgtcagat ccagtgcatc 360
aatggcccga atgacgagag attgctgctc ggcactgaaa gtctgcagcg tgcttgtttc 420
ggagttatct ctacagctgc accgaatgac gaacaagccc gactatccat gtccgacgag 480
cccggaccga cgtgatccga atgctgtcta acacttacca agacaatcat ggggtatctc 540
tgaattatga ttgtgttgct caagcagcta atcccaactc ttaatcacgt caagtgccat 600
gcttacctgt attatcatct gtcatcaaca taccaactcc tatatatagg ggattgtgct 660
tcattacctc tccagtaaca actacttgga gttcaaacca catttcaact tactctttta 720
gttcattcaa catgacccca tcaatgatgg aggtattcgt ttttggggac caaagcacac 780
gctttgcccc tccactgaaa gacctactcc tcaaaggcaa cagtccttac ttgacacatt 840
ttgttaaaca agttcacgca cttcttagaa gggagatatc atccttgccg gcagttcaac 900
agaagctttt cccaaacttt gccgacattc aggaactcgt ctccaagtca gattggggca 960
gtggtaaccc tgctttgaca agcgctttag catgctttta ccatctttgc agtttcattc 1020
aggagtatca taaccaagta cccatagatc tctcctgacc atccagcttt tacgatggac 1080
aaggtcgtac ctttccttcg gagaacagtc gcattattgg actttgcgtt ggttcactcg 1140
ctgctactgc tgtcagttgc tccacatcac tgagtgaatt ggtatcagct ggtgtagatg 1200
ctgttcgtgt ggcattgcac gtcggactac gggtatggcg aactacctcc cttttcgatg 1260
taccagacag gccctccgcc acttggttca taattgtgcc cgaggcagta ctaccaagag 1320
aatctgcgca agaccgactt gactcattca tcattgaaat ggtaagcaat tctttgtcct 1380
tggagctctc ataaaacata ttcactcttc tttagggact tgctcgatca tcagttcctt 1440
acatcagctc ggtcgcacat cacaacatga ccatcagtgg tccaccatcc gtcctcgaaa 1500
agttcattca cagtatatca acatcaccga aagattctct tccagtgccg atctatgctc 1560
cgtaccacgc cagccatctt tacagcatgg atgatgtaga cgaggtcctt agcctgtctg 1620
caccttcttt tgcatcagag tccatcattc cactcatttc aagctcctcg ggtgacgagt 1680
tacagccact caagtatgca gatctactcc gctgctgtgt tagtgatatg ctcatacagc 1740
cactggatct taccaaggtc tcacaagcag tggcccagct tctcgaggtt agctcatcta 1800
cacgtgccat aataaagcct atagcaacca gcgtctccaa cagtctagtg tctgttttgg 1860
agccgacgct agcagaacga tgcgccgtgg acaacagcat ggggcccaaa gcctcgacca 1920
gccactcatc agcagagaca caaaccgagt catcaagcaa gaactccaaa attgcgattg 1980

ttgctatgtc tggtcgcttt ccagacgcag ctgacttgag tgaattctgg gatcttctct 2040
acgaaggtcg cgatgttcat cgacaaattc ccgaggaccg attcaacgca gagctccatt 2100
acgacgctac tgggcgacgt aagaacacca gcaaggtcat gaatggctgc ttcatcaagg 2160
aaccaggact gttcgacgct aggttcttca acatgtctcc aaaggaagct gagcagtcgg 2220
atcctggcca gcgaatggcc ctcgaaacag cttacgaggc gcttgagatg gctagtatcg 2280
taccagacag aacaccttcg acacagagag atcgtgttgg tgtgttctac ggcatgacta 2340
gcgatgattg gagagaggtc aacagtgggc agaatgtcga cacttatttt attcctggta 2400
agttgtttgt tcgcgtttca cgacaatact gattgcagac tacaggtggc aacagagcgt 2460
tcactcctgg tcgactcaac tacttcttca agttcagtgg gcctagcgct agtgttgata 2520
cggcttgctc ctccagtctc gttggcttgc acttggcttg taattccctc tggagaaatg 2580
attgcgatac agctattgcg ggcggaacca atgtcatgac taaccctgac aacttcgctg 2640
gtttggaccg aggccacttc ctatctagaa ccggtaagtc tctcacagat cttggaagct 2700
tccttctttt tagtgttttt actgaccttg ttccaggcaa ctgcaacacc tttgacgatg 2760
gagcagacgg atactgtcga gctgatggcg tcggaaccat catcctcaag cggcttgagg 2820
acgccgaagc tgacaatgac cctattctcg gtgtcattct gggcgcttac acaaaccact 2880
cagccgaagc agtatccatc actcgaccac atgccggagc tcaagagtac atcttctcca 2940
aactcctccg tgagtcgggc accgatccct acaacgttag ctacatcgag atgcacggca 3000
caggcactca agccggcgac gcaaccgaga tgacatccgt cctcaagacg tttgctccta 3060
ccagcggctt cggcggtcga ttgcctcacc aaaaccttca cttgggttca gtcaaggcca 3120
atgtcgggca cggtgaatcc gcatctggta tcattgctct gatcaagacg ctgcttatga 3180
tggagaagaa catgatcccg ccgcattgtg gtatcaagac aaagatcaat caccattttc 3240
ctacggatct cactcagcgc aatgtccata tcgccaaagt tccgacatct tggacaagat 3300
cgggtcaagc caatccacgc attgctttcg tcaataactt ctctgccgct ggtggtaact 3360
ctgctgtcct actgcaagat gctcctcagc catcggtagt ttcggatgtt acagaccctc 3420
gcacatccca tgttgtcact atgtccgctc gatcagcaga ttccctcagg aagaacctcg 3480
ccaatctcaa ggagcttgta gaaggccaag gtgactcgga ggtcggcttc ctgagcaagc 3540
tgtcctacac aaccaccgcc aggcgcatgc atcatcaatt ccgagcttcg gtcacagcac 3600
agactcgtga acagctgctg aagggccttg attccgccat tgaacgccag gatgtgaaga 3660
ggatccccgc cgccgcgccc tctgtcggct ttgtgtttag cggccaaggc gcccaatacc 3720
gtggtatggg caaggagtac tttacatctt tcacagcctt ccgctctgag atcatgtctt 3780
acgacagtat cgcccaagcc caaggcttcc cgtcaatcct cccactgatc cgaggagagg 3840
tggaagctga ctcgttgagt cctgttgaga tccagctggg tctcacttgc ctgcagatgg 3900
cactggccaa gctatggaag tcattcggtg ttgagccagg ctttgttctc ggacacagct 3960
taggccacta tgctgcttta cacgtcgctg gtgttctgtc cgccaatgat accatttacc 4020
tcactggcat cagagcgcag ctgctcgtgg ataagtgcca ggcaggaacc cactcaatgc 4080
tggcagtgag ggcatcctta ctacagatcc aacagttcct cgatgccaac attcacgagg 4140
ttgcatgtgt caatggttca cgagaagtcg tcatcagtgg acgcgttgcc gacattgacc 4200
agctggttgg ccttttgtcg gctgacaaca tcaaggcgac ccgcgtcaag gtgccatttg 4260
ccttccactc agcgcaggtt gaccccattc tctccgactt ggatacagcg gcgtcgcgcg 4320
tcaccttcca ctccctccag attcctgttc tttgtgccct tgacagctct gtcatcagcc 4380
ctggaaacca tggtgtcatt ggtccccttc atctacagcg acattgtcgt gagacagtca 4440
actttgaggg tgctctacat gctgcggagc acgagaagat catcaacaag acatcaactc 4500
tatggatcga gattggtccc catgttgtct gctctacctt cctcaagtcc agccttggtc 4560
caagcacacc tgctatcgca tcgcttcgcc gaaatgacga ttgctggaag gtgttggctg 4620
atggtttgag cagtctctac agcagcgggt tgacaattga cttgaacgag tatcatcgcg 4680
acttcaaggc ctctcaccag gtacttcgtc tgccttgtta cagctgggag cacaagaatt 4740
actggataca gtacaagtac gattggtcct tggctaaagg tgatcctcca attgccccta 4800
acagctcggt tgaagcagtc tcagctttat caacaccctc ggtccagaag attctacagg 4860
agacttccct tgatcaggta ttgactatcg ttgctgagac agatctcgcg agccctctat 4920

tgtcagaggt tgcccaaggt catcgggtca acggtgtcaa agtctgcaca tcttccgtgt 4980
acgctgatgt tggcttgacg ctgggtaagt acattttgga caactaccgc accgacttag 5040
agggttatgc ggtcgatgtt cacggtattg aggtccacaa gccacttctt ctcaaagaag 5100
acatgaacgg aacgccccag gctacaccgt tccgcatcga agtgcgatac ccaatccaga 5160
gcaccacggc gctgatgagc atctccacca ctggccccaa cggtcagcac atcaagcatg 5220
ctaactgcga acttcgactc gagcatccgt cgcaatggga agcggagtgg gatcgccaag 5280
cctacctcat caatcgcagc gtcaactgcc ttctgcagcg atcagcacaa ggtttggaca 5340
gcatgttggc aaccggaatg gtctacaagg tcttctcctc cctcgtcgac tatgccgatg 5400
gctacaaggg tctgcaggag gttgtcttgc acagccaaga gctcgagggc acagcaaaag 5460
tgcgcttcca aactccctcg ggaggtttcg tctgcaatcc catgtggatt gacagctgtg 5520
gtcagacgac cggcttcatg atgaactgtc atcagactac gcccaatgac tacgtctacg 5580
tcaatcatgg ctggaagtcg atgagattgg ccaaggcgtt ccgtgaagat ggtacctatc 5640
ggacttatat ccggatgagg cccattgata gcaccaagtt cgctggtgac ttgtacattc 5700
ttgatgagga tgacactgtg gttggtgttt atggagacat aacaatgagt aacacgaatc 5760
tttgtcaaca acaatgctaa cgtaaagaac agttccaagg tttgccgcga cgagttctca 5820
acacagtctt gccatctgcc aacgcggttc cagttgatgc tcccatgggt cgacgggacg 5880
tgcctccatc aagaatggat gtgcctcccg tcaggtccgg tgaagggcca cccacttcag 5940
cacccacgca gcaagctatc gctctgccgt tcgcagccga tacatccatg gactcccgat 6000
tgagacctct tcttcgcatc ttgtcagaag agatcggtct cggtcttgac gttctttcgg 6060
acgatgaact cgactttgcg gaccacggtg tcgactcact cctctcattg accatcactg 6120
gtcgcatgcg tgaggaattg ggtctcgacg ttgaatctac agcattcatg aactgtccca 6180
ctttgggcag ctttaaattg ttcctaggac ttgtcgatca ggacaataag ggcagcagcg 6240
gcagtgatgg cagtggtagg agcagtccag caccgggtac cgagtctggc gctactacac 6300
cacctatgag cgaagaggac caggacaaga tagtcagcag tcactcgctt caccagttcc 6360
aagccagttc gacgcttcta cagggcagtc ccagtaaagc tcgctcgact ttgttcttgc 6420
taccagatgg ctcgggatct gccacatcct acgcttccct tcccccgatc tctccagacg 6480
gagatgttgc tgtctacggg ttgaactgtc catggctgaa ggactctagt tacctcgtcg 6540
agtttggact caagggcttg acagagctct atgtcaacga gatactccgt cgcaagccac 6600
agggtcctta caatttggga ggatggtcag ccggtggcat ttgcgcttat gaagctgccc 6660
tgatcctcac cagagcagga caccaagtcg atcgccttat cttgattgac tctcccaatc 6720
ccgttggtct tgagaagcta cctcctcgct tgtacgattt cctcaattcg cagaatgtct 6780
ttggatcaga caacccgcac agcactgctg gaacaagcgt caaagctcca gaatggcttc 6840
ttgcacattt cctggccttc attgacgctc tggatgctta tgtcgcagtg ccttgggact 6900
ctggtctagt cggtctagca tcaccgctcc ctgcaccgcc gcagacatac atgctgtggg 6960
cagaagacgg agtttgcaaa gactctgata gtgctcgtcc cgagtaccgt gacgatgacc 7020
cacgcgagat gagatggctg ttggagaaca gaacaaactt tggtccgaat ggttgggagg 7080
cgctacttgg tggtaaagag ggtttgttca tggatcggat tgcggaagcg aatcatttta 7140
gtatgttgaa gagaggacgg aatgcggaat atgtctctgc attcctggct cgggccttgg 7200
acaattagcg atggagaggg ctgtttgtgt atttatgttt attcttcttt gatgtttgta 7260
tttctttgtc tttctcatca ttcagataga agcagcagat tgccttcatt ctttataaat 7320
cactacagaa ataagtttg 7339

<210> 4
<211> 6608
<212> DNA
<213> Fusarium graminearum

<400> 4
atgaccccat caatgatgga ggtattcgtt tttggggacc aaagcacacg ctttgcccct 60
ccactgaaag acctactcct caaaggcaac agtccttact tgacacattt tgttaaacaa 120
gttcacgcac ttcttagaag ggagatatca tccttgccgg cagttcaaca gaagcttttc 180

ccaaactttg ccgacattca ggaactcgtc tccaagtcag attggggcag tggtaaccct 240
gctttgacaa gcgctttagc atgcttttac catctttgca gtttcattca ggagtatcat 300
aaccaagtac ccatagatct ctcctgacca tccagctttt acgatggaca aggtcgtacc 360
tttccttcgg agaacagtcg cattattgga ctttgcgttg gttcactcgc tgctactgct 420
gtcagttgct ccacatcact gagtgaattg gtatcagctg gtgtagatgc tgttcgtgtg 480
gcattgcacg tcggactacg ggtatggcga actacctccc ttttcgatgt accagacagg 540
ccctccgcca cttggttcat aattgtgccc gaggcagtac taccaagaga atctgcgcaa 600
gaccgacttg actcattcat cattgaaatg gtaagcaatt ctttgtcctt ggagctctca 660
taaaacatat tcactcttct ttagggactt gctcgatcat cagttcctta catcagctcg 720
gtcgcacatc acaacatgac catcagtggt ccaccatccg tcctcgaaaa gttcattcac 780
agtatatcaa catcaccgaa agattctctt ccagtgccga tctatgctcc gtaccacgcc 840
agccatcttt acagcatgga tgatgtagac gaggtcctta gcctgtctgc accttctttt 900
gcatcagagt ccatcattcc actcatttca agctcctcgg gtgacgagtt acagccactc 960
aagtatgcag atctactccg ctgctgtgtt agtgatatgc tcatacagcc actggatctt 1020
accaaggtct cacaagcagt ggcccagctt ctcgaggtta gctcatctac acgtgccata 1080
ataaagccta tagcaaccag cgtctccaac agtctagtgt ctgttttgga gccgacgcta 1140
gcagaacgat gcgccgtgga caacagcatg gggcccaaag cctcgaccag ccactcatca 1200
gcagagacac aaaccgagtc atcaagcaag aactccaaaa ttgcgattgt tgctatgtct 1260
ggtcgctttc cagacgcagc tgacttgagt gaattctggg atcttctcta cgaaggtcgc 1320
gatgttcatc gacaaattcc cgaggaccga ttcaacgcag agctccatta cgacgctact 1380
gggcgacgta agaacaccag caaggtcatg aatggctgct tcatcaagga accaggactg 1440
ttcgacgcta ggttcttcaa catgtctcca aaggaagctg agcagtcgga tcctggccag 1500
cgaatggccc tcgaaacagc ttacgaggcg cttgagatgg ctagtatcgt accagacaga 1560
acaccttcga cacagagaga tcgtgttggt gtgttctacg gcatgactag cgatgattgg 1620
agagaggtca acagtgggca gaatgtcgac acttatttta ttcctggtaa gttgtttgtt 1680
cgcgtttcac gacaatactg attgcagact acaggtggca acagagcgtt cactcctggt 1740
cgactcaact acttcttcaa gttcagtggg cctagcgcta gtgttgatac ggcttgctcc 1800
tccagtctcg ttggcttgca cttggcttgt aattccctct ggagaaatga ttgcgataca 1860
gctattgcgg gcggaaccaa tgtcatgact aaccctgaca acttcgctgg tttggaccga 1920
ggccacttcc tatctagaac cggtaagtct ctcacagatc ttggaagctt ccttcttttt 1980
agtgttttta ctgaccttgt tccaggcaac tgcaacacct ttgacgatgg agcagacgga 2040
tactgtcgag ctgatggcgt cggaaccatc atcctcaagc ggcttgagga cgccgaagct 2100
gacaatgacc ctattctcgg tgtcattctg ggcgcttaca caaaccactc agccgaagca 2160
gtatccatca ctcgaccaca tgccggagct caagagtaca tcttctccaa actcctccgt 2220
gagtcgggca ccgatcccta caacgttagc tacatcgaga tgcacggcac aggcactcaa 2280
gccggcgacg caaccgagat gacatccgtc ctcaagacgt ttgctcctac cagcggcttc 2340
ggcggtcgat tgcctcacca aaaccttcac ttgggttcag tcaaggccaa tgtcgggcac 2400
ggtgaatccg catctggtat cattgctctg atcaagacgc tgcttatgat ggagaagaac 2460
atgatcccgc cgcattgtgg tatcaagaca aagatcaatc accattttcc tacggatctc 2520
actcagcgca atgtccatat cgccaaagtt ccgacatctt ggacaagatc gggtcaagcc 2580
aatccacgca ttgctttcgt caataacttc tctgccgctg gtggtaactc tgctgtccta 2640
ctgcaagatg ctcctcagcc atcggtagtt tcggatgtta cagaccctcg cacatcccat 2700
gttgtcacta tgtccgctcg atcagcagat tccctcagga agaacctcgc caatctcaag 2760
gagcttgtag aaggccaagg tgactcggag gtcggcttcc tgagcaagct gtcctacaca 2820
accaccgcca ggcgcatgca tcatcaattc cgagcttcgg tcacagcaca gactcgtgaa 2880
cagctgctga agggccttga ttccgccatt gaacgccagg atgtgaagag gatccccgcc 2940
gccgcgccct ctgtcggctt tgtgtttagc ggccaaggcg cccaataccg tggtatgggc 3000
aaggagtact ttacatcttt cacagccttc cgctctgaga tcatgtctta cgacagtatc 3060
gcccaagccc aaggcttccc gtcaatcctc ccactgatcc gaggagaggt ggaagctgac 3120

tcgttgagtc ctgttgagat ccagctgggt ctcacttgcc tgcagatggc actggccaag 3180
ctatggaagt cattcggtgt tgagccaggc tttgttctcg gacacagctt aggccactat 3240
gctgctttac acgtcgctgg tgttctgtcc gccaatgata ccatttacct cactggcatc 3300
agagcgcagc tgctcgtgga taagtgccag gcaggaaccc actcaatgct ggcagtgagg 3360
gcatccttac tacagatcca acagttcctc gatgccaaca ttcacgaggt tgcatgtgtc 3420
aatggttcac gagaagtcgt catcagtgga cgcgttgccg acattgacca gctggttggc 3480
cttttgtcgg ctgacaacat caaggcgacc cgcgtcaagg tgccatttgc cttccactca 3540
gcgcaggttg accccattct ctccgacttg gatacagcgg cgtcgcgcgt caccttccac 3600
tccctccaga ttcctgttct ttgtgccctt gacagctctg tcatcagccc tggaaaccat 3660
ggtgtcattg gtccccttca tctacagcga cattgtcgtg agacagtcaa ctttgagggt 3720
gctctacatg ctgcggagca cgagaagatc atcaacaaga catcaactct atggatcgag 3780
attggtcccc atgttgtctg ctctaccttc ctcaagtcca gccttggtcc aagcacacct 3840
gctatcgcat cgcttcgccg aaatgacgat tgctggaagg tgttggctga tggtttgagc 3900
agtctctaca gcagcgggtt gacaattgac ttgaacgagt atcatcgcga cttcaaggcc 3960
tctcaccagg tacttcgtct gccttgttac agctgggagc acaagaatta ctggatacag 4020
tacaagtacg attggtcctt ggctaaaggt gatcctccaa ttgcccctaa cagctcggtt 4080
gaagcagtct cagctttatc aacaccctcg gtccagaaga ttctacagga gacttccctt 4140
gatcaggtat tgactatcgt tgctgagaca gatctcgcga gccctctatt gtcagaggtt 4200
gcccaaggtc atcgggtcaa cggtgtcaaa gtctgcacat cttccgtgta cgctgatgtt 4260
ggcttgacgc tgggtaagta cattttggac aactaccgca ccgacttaga gggttatgcg 4320
gtcgatgttc acggtattga ggtccacaag ccacttcttc tcaaagaaga catgaacgga 4380
acgccccagg ctacaccgtt ccgcatcgaa gtgcgatacc caatccagag caccacggcg 4440
ctgatgagca tctccaccac tggccccaac ggtcagcaca tcaagcatgc taactgcgaa 4500
cttcgactcg agcatccgtc gcaatgggaa gcggagtggg atcgccaagc ctacctcatc 4560
aatcgcagcg tcaactgcct tctgcagcga tcagcacaag gtttggacag catgttggca 4620
accggaatgg tctacaaggt cttctcctcc ctcgtcgact atgccgatgg ctacaagggt 4680
ctgcaggagg ttgtcttgca cagccaagag ctcgagggca cagcaaaagt gcgcttccaa 4740
actccctcgg gaggtttcgt ctgcaatccc atgtggattg acagctgtgg tcagacgacc 4800
ggcttcatga tgaactgtca tcagactacg cccaatgact acgtctacgt caatcatggc 4860
tggaagtcga tgagattggc caaggcgttc cgtgaagatg gtacctatcg gacttatatc 4920
cggatgaggc ccattgatag caccaagttc gctggtgact tgtacattct tgatgaggat 4980
gacactgtgg ttggtgttta tggagacata acaatgagta acacgaatct ttgtcaacaa 5040
caatgctaac gtaaagaaca gttccaaggt ttgccgcgac gagttctcaa cacagtcttg 5100
ccatctgcca acgcggttcc agttgatgct cccatgggtc gacgggacgt gcctccatca 5160
agaatggatg tgcctcccgt caggtccggt gaagggccac ccacttcagc acccacgcag 5220
caagctatcg ctctgccgtt cgcagccgat acatccatgg actcccgatt gagacctctt 5280
cttcgcatct tgtcagaaga gatcggtctc ggtcttgacg ttctttcgga cgatgaactc 5340
gactttgcgg accacggtgt cgactcactc ctctcattga ccatcactgg tcgcatgcgt 5400
gaggaattgg gtctcgacgt tgaatctaca gcattcatga actgtcccac tttgggcagc 5460
tttaaattgt tcctaggact tgtcgatcag gacaataagg gcagcagcgg cagtgatggc 5520
agtggtagga gcagtccagc accgggtacc gagtctggcg ctactacacc acctatgagc 5580
gaagaggacc aggacaagat agtcagcagt cactcgcttc accagttcca agccagttcg 5640
acgcttctac agggcagtcc cagtaaagct cgctcgactt tgttcttgct accagatggc 5700
tcgggatctg ccacatccta cgcttccctt cccccgatct ctccagacgg agatgttgct 5760
gtctacgggt tgaactgtcc atggctgaag gactctagtt acctcgtcga gtttggactc 5820
aagggcttga cagagctcta tgtcaacgag atactccgtc gcaagccaca gggtccttac 5880
aatttgggag gatggtcagc cggtggcatt tgcgcttatg aagctgccct gatcctcacc 5940
agagcaggac accaagtcga tcgccttatc ttgattgact ctcccaatcc cgttggtctt 6000
gagaagctac ctcctcgctt gtacgatttc ctcaattcgc agaatgtctt tggatcagac 6060

aacccgcaca gcactgctgg aacaagcgtc aaagctccag aatggcttct tgcacatttc 6120
ctggccttca ttgacgctct ggatgcttat gtcgcagtgc cttgggactc tggtctagtc 6180
ggtctagcat caccgctccc tgcaccgccg cagacataca tgctgtgggc agaagacgga 6240
gtttgcaaag actctgatag tgctcgtccc gagtaccgtg acgatgaccc acgcgagatg 6300
agatggctgt tggagaacag aacaaacttt ggtccgaatg gttgggaggc gctacttggt 6360
ggtaaagagg gtttgttcat ggatcggatt gcggaagcga atcattttag tatgttgaag 6420
agaggacgga atgcggaata tgtctctgca ttcctggctc gggccttgga caattagcga 6480
tggagagggc tgtttgtgta tttatgttta ttcttctttg atgtttgtat ttctttgtct 6540
ttctcatcat tcagatagaa gcagcagatt gccttcattc tttataaatc actacagaaa 6600
taagtttg 6608

<210> 5
<211> 6219
<212> DNA
<213> Fusarium graminearum

<220>
<221> CDS
<222> (1)..(6216)

<400> 5

```
atg acc cca tca atg atg gag gta ttc gtt ttt ggg gac caa agc aca   48
Met Thr Pro Ser Met Met Glu Val Phe Val Phe Gly Asp Gln Ser Thr
  1               5                  10                  15

cgc ttt gcc cct cca ctg aaa gac cta ctc ctc aaa ggc aac agt cct   96
Arg Phe Ala Pro Pro Leu Lys Asp Leu Leu Leu Lys Gly Asn Ser Pro
             20                  25                  30

tac ttg aca cat ttt gtt aaa caa gtt cac gca ctt ctt aga agg gag   144
Tyr Leu Thr His Phe Val Lys Gln Val His Ala Leu Leu Arg Arg Glu
         35                  40                  45

ata tca tcc ttg ccg gca gtt caa cag aag ctt ttc cca aac ttt gcc   192
Ile Ser Ser Leu Pro Ala Val Gln Gln Lys Leu Phe Pro Asn Phe Ala
     50                  55                  60

gac att cag gaa ctc gtc tcc aag tca gat tgg ggc agt ggt aac cct   240
Asp Ile Gln Glu Leu Val Ser Lys Ser Asp Trp Gly Ser Gly Asn Pro
 65                  70                  75                  80

gct ttg aca agc gct tta gca tgc ttt tac cat ctt tgc agt ttc att   288
Ala Leu Thr Ser Ala Leu Ala Cys Phe Tyr His Leu Cys Ser Phe Ile
                 85                  90                  95

cac ttt tac gat gga caa ggt cgt acc ttt cct tcg gag aac agt cgc   336
His Phe Tyr Asp Gly Gln Gly Arg Thr Phe Pro Ser Glu Asn Ser Arg
            100                 105                 110

att att gga ctt tgc gtt ggt tca ctc gct gct act gct gtc agt tgc   384
Ile Ile Gly Leu Cys Val Gly Ser Leu Ala Ala Thr Ala Val Ser Cys
        115                 120                 125
```

```
tcc aca tca ctg agt gaa ttg gta tca gct ggt gta gat gct gtt cgt   432
Ser Thr Ser Leu Ser Glu Leu Val Ser Ala Gly Val Asp Ala Val Arg
    130                 135                 140

gtg gca ttg cac gtc gga cta cgg gta tgg cga act acc tcc ctt ttc   480
Val Ala Leu His Val Gly Leu Arg Val Trp Arg Thr Thr Ser Leu Phe
145                 150                 155                 160

gat gta cca gac agg ccc tcc gcc act tgg ttc ata att gtg ccc gag   528
Asp Val Pro Asp Arg Pro Ser Ala Thr Trp Phe Ile Ile Val Pro Glu
                165                 170                 175

gca gta cta cca aga gaa tct gcg caa gac cga ctt gac tca ttc atc   576
Ala Val Leu Pro Arg Glu Ser Ala Gln Asp Arg Leu Asp Ser Phe Ile
            180                 185                 190

att gaa atg gga ctt gct cga tca tca gtt cct tac atc agc tcg gtc   624
Ile Glu Met Gly Leu Ala Arg Ser Ser Val Pro Tyr Ile Ser Ser Val
        195                 200                 205

gca cat cac aac atg acc atc agt ggt cca cca tcc gtc ctc gaa aag   672
Ala His His Asn Met Thr Ile Ser Gly Pro Pro Ser Val Leu Glu Lys
    210                 215                 220

ttc att cac agt ata tca aca tca ccg aaa gat tct ctt cca gtg ccg   720
Phe Ile His Ser Ile Ser Thr Ser Pro Lys Asp Ser Leu Pro Val Pro
225                 230                 235                 240

atc tat gct ccg tac cac gcc agc cat ctt tac agc atg gat gat gta   768
Ile Tyr Ala Pro Tyr His Ala Ser His Leu Tyr Ser Met Asp Asp Val
                245                 250                 255

gac gag gtc ctt agc ctg tct gca cct tct ttt gca tca gag tcc atc   816
Asp Glu Val Leu Ser Leu Ser Ala Pro Ser Phe Ala Ser Glu Ser Ile
            260                 265                 270

att cca ctc att tca agc tcc tcg ggt gac gag tta cag cca ctc aag   864
Ile Pro Leu Ile Ser Ser Ser Ser Gly Asp Glu Leu Gln Pro Leu Lys
        275                 280                 285

tat gca gat cta ctc cgc tgc tgt gtt agt gat atg ctc ata cag cca   912
Tyr Ala Asp Leu Leu Arg Cys Cys Val Ser Asp Met Leu Ile Gln Pro
    290                 295                 300

ctg gat ctt acc aag gtc tca caa gca gtg gcc cag ctt ctc gag gtt   960
Leu Asp Leu Thr Lys Val Ser Gln Ala Val Ala Gln Leu Leu Glu Val
305                 310                 315                 320

agc tca tct aca cgt gcc ata ata aag cct ata gca acc agc gtc tcc  1008
Ser Ser Ser Thr Arg Ala Ile Ile Lys Pro Ile Ala Thr Ser Val Ser
                325                 330                 335

aac agt cta gtg tct gtt ttg gag ccg acg cta gca gaa cga tgc gcc  1056
Asn Ser Leu Val Ser Val Leu Glu Pro Thr Leu Ala Glu Arg Cys Ala
            340                 345                 350
```

```
gtg gac aac agc atg ggg ccc aaa gcc tcg acc agc cac tca tca gca   1104
Val Asp Asn Ser Met Gly Pro Lys Ala Ser Thr Ser His Ser Ser Ala
        355                 360                 365

gag aca caa acc gag tca tca agc aag aac tcc aaa att gcg att gtt   1152
Glu Thr Gln Thr Glu Ser Ser Ser Lys Asn Ser Lys Ile Ala Ile Val
    370                 375                 380

gct atg tct ggt cgc ttt cca gac gca gct gac ttg agt gaa ttc tgg   1200
Ala Met Ser Gly Arg Phe Pro Asp Ala Ala Asp Leu Ser Glu Phe Trp
385                 390                 395                 400

gat ctt ctc tac gaa ggt cgc gat gtt cat cga caa att ccc gag gac   1248
Asp Leu Leu Tyr Glu Gly Arg Asp Val His Arg Gln Ile Pro Glu Asp
                405                 410                 415

cga ttc aac gca gag ctc cat tac gac gct act ggg cga cgt aag aac   1296
Arg Phe Asn Ala Glu Leu His Tyr Asp Ala Thr Gly Arg Arg Lys Asn
            420                 425                 430

acc agc aag gtc atg aat ggc tgc ttc atc aag gaa cca gga ctg ttc   1344
Thr Ser Lys Val Met Asn Gly Cys Phe Ile Lys Glu Pro Gly Leu Phe
        435                 440                 445

gac gct agg ttc ttc aac atg tct cca aag gaa gct gag cag tcg gat   1392
Asp Ala Arg Phe Phe Asn Met Ser Pro Lys Glu Ala Glu Gln Ser Asp
    450                 455                 460

cct ggc cag cga atg gcc ctc gaa aca gct tac gag gcg ctt gag atg   1440
Pro Gly Gln Arg Met Ala Leu Glu Thr Ala Tyr Glu Ala Leu Glu Met
465                 470                 475                 480

gct agt atc gta cca gac aga aca cct tcg aca cag aga gat cgt gtt   1488
Ala Ser Ile Val Pro Asp Arg Thr Pro Ser Thr Gln Arg Asp Arg Val
                485                 490                 495

ggt gtg ttc tac ggc atg act agc gat gat tgg aga gag gtc aac agt   1536
Gly Val Phe Tyr Gly Met Thr Ser Asp Asp Trp Arg Glu Val Asn Ser
            500                 505                 510

ggg cag aat gtc gac act tat ttt att cct ggt ggc aac aga gcg ttc   1584
Gly Gln Asn Val Asp Thr Tyr Phe Ile Pro Gly Gly Asn Arg Ala Phe
        515                 520                 525

act cct ggt cga ctc aac tac ttc ttc aag ttc agt ggg cct agc gct   1632
Thr Pro Gly Arg Leu Asn Tyr Phe Phe Lys Phe Ser Gly Pro Ser Ala
    530                 535                 540

agt gtt gat acg gct tgc tcc tcc agt ctc gtt ggc ttg cac ttg gct   1680
Ser Val Asp Thr Ala Cys Ser Ser Ser Leu Val Gly Leu His Leu Ala
545                 550                 555                 560

tgt aat tcc ctc tgg aga aat gat tgc gat aca gct att gcg ggc gga   1728
Cys Asn Ser Leu Trp Arg Asn Asp Cys Asp Thr Ala Ile Ala Gly Gly
                565                 570                 575
```

```
acc aat gtc atg act aac cct gac aac ttc gct ggt ttg gac cga ggc   1776
Thr Asn Val Met Thr Asn Pro Asp Asn Phe Ala Gly Leu Asp Arg Gly
            580                 585                 590

cac ttc cta tct aga acc ggc aac tgc aac acc ttt gac gat gga gca   1824
His Phe Leu Ser Arg Thr Gly Asn Cys Asn Thr Phe Asp Asp Gly Ala
        595                 600                 605

gac gga tac tgt cga gct gat ggc gtc gga acc atc atc ctc aag cgg   1872
Asp Gly Tyr Cys Arg Ala Asp Gly Val Gly Thr Ile Ile Leu Lys Arg
    610                 615                 620

ctt gag gac gcc gaa gct gac aat gac cct att ctc ggt gtc att ctg   1920
Leu Glu Asp Ala Glu Ala Asp Asn Asp Pro Ile Leu Gly Val Ile Leu
625                 630                 635                 640

ggc gct tac aca aac cac tca gcc gaa gca gta tcc atc act cga cca   1968
Gly Ala Tyr Thr Asn His Ser Ala Glu Ala Val Ser Ile Thr Arg Pro
                645                 650                 655

cat gcc gga gct caa gag tac atc ttc tcc aaa ctc ctc cgt gag tcg   2016
His Ala Gly Ala Gln Glu Tyr Ile Phe Ser Lys Leu Leu Arg Glu Ser
            660                 665                 670

ggc acc gat ccc tac aac gtt agc tac atc gag atg cac ggc aca ggc   2064
Gly Thr Asp Pro Tyr Asn Val Ser Tyr Ile Glu Met His Gly Thr Gly
        675                 680                 685

act caa gcc ggc gac gca acc gag atg aca tcc gtc ctc aag acg ttt   2112
Thr Gln Ala Gly Asp Ala Thr Glu Met Thr Ser Val Leu Lys Thr Phe
    690                 695                 700

gct cct acc agc ggc ttc ggc ggt cga ttg cct cac caa aac ctt cac   2160
Ala Pro Thr Ser Gly Phe Gly Gly Arg Leu Pro His Gln Asn Leu His
705                 710                 715                 720

ttg ggt tca gtc aag gcc aat gtc ggg cac ggt gaa tcc gca tct ggt   2208
Leu Gly Ser Val Lys Ala Asn Val Gly His Gly Glu Ser Ala Ser Gly
                725                 730                 735

atc att gct ctg atc aag acg ctg ctt atg atg gag aag aac atg atc   2256
Ile Ile Ala Leu Ile Lys Thr Leu Leu Met Met Glu Lys Asn Met Ile
            740                 745                 750

ccg ccg cat tgt ggt atc aag aca aag atc aat cac cat ttt cct acg   2304
Pro Pro His Cys Gly Ile Lys Thr Lys Ile Asn His His Phe Pro Thr
        755                 760                 765

gat ctc act cag cgc aat gtc cat atc gcc aaa gtt ccg aca tct tgg   2352
Asp Leu Thr Gln Arg Asn Val His Ile Ala Lys Val Pro Thr Ser Trp
    770                 775                 780

aca aga tcg ggt caa gcc aat cca cgc att gct ttc gtc aat aac ttc   2400
Thr Arg Ser Gly Gln Ala Asn Pro Arg Ile Ala Phe Val Asn Asn Phe
785                 790                 795                 800
```

```
tct gcc gct ggt ggt aac tct gct gtc cta ctg caa gat gct cct cag   2448
Ser Ala Ala Gly Gly Asn Ser Ala Val Leu Leu Gln Asp Ala Pro Gln
                805                 810                 815

cca tcg gta gtt tcg gat gtt aca gac cct cgc aca tcc cat gtt gtc   2496
Pro Ser Val Val Ser Asp Val Thr Asp Pro Arg Thr Ser His Val Val
            820                 825                 830

act atg tcc gct cga tca gca gat tcc ctc agg aag aac ctc gcc aat   2544
Thr Met Ser Ala Arg Ser Ala Asp Ser Leu Arg Lys Asn Leu Ala Asn
        835                 840                 845

ctc aag gag ctt gta gaa ggc caa ggt gac tcg gag gtc ggc ttc ctg   2592
Leu Lys Glu Leu Val Glu Gly Gln Gly Asp Ser Glu Val Gly Phe Leu
    850                 855                 860

agc aag ctg tcc tac aca acc acc gcc agg cgc atg cat cat caa ttc   2640
Ser Lys Leu Ser Tyr Thr Thr Thr Ala Arg Arg Met His His Gln Phe
865                 870                 875                 880

cga gct tcg gtc aca gca cag act cgt gaa cag ctg ctg aag ggc ctt   2688
Arg Ala Ser Val Thr Ala Gln Thr Arg Glu Gln Leu Leu Lys Gly Leu
                885                 890                 895

gat tcc gcc att gaa cgc cag gat gtg aag agg atc ccc gcc gcc gcg   2736
Asp Ser Ala Ile Glu Arg Gln Asp Val Lys Arg Ile Pro Ala Ala Ala
            900                 905                 910

ccc tct gtc ggc ttt gtg ttt agc ggc caa ggc gcc caa tac cgt ggt   2784
Pro Ser Val Gly Phe Val Phe Ser Gly Gln Gly Ala Gln Tyr Arg Gly
        915                 920                 925

atg ggc aag gag tac ttt aca tct ttc aca gcc ttc cgc tct gag atc   2832
Met Gly Lys Glu Tyr Phe Thr Ser Phe Thr Ala Phe Arg Ser Glu Ile
    930                 935                 940

atg tct tac gac agt atc gcc caa gcc caa ggc ttc ccg tca atc ctc   2880
Met Ser Tyr Asp Ser Ile Ala Gln Ala Gln Gly Phe Pro Ser Ile Leu
945                 950                 955                 960

cca ctg atc cga gga gag gtg gaa gct gac tcg ttg agt cct gtt gag   2928
Pro Leu Ile Arg Gly Glu Val Glu Ala Asp Ser Leu Ser Pro Val Glu
                965                 970                 975

atc cag ctg ggt ctc act tgc ctg cag atg gca ctg gcc aag cta tgg   2976
Ile Gln Leu Gly Leu Thr Cys Leu Gln Met Ala Leu Ala Lys Leu Trp
            980                 985                 990

aag tca ttc ggt gtt gag cca ggc ttt gtt ctc gga cac agc tta ggc   3024
Lys Ser Phe Gly Val Glu Pro Gly Phe Val Leu Gly His Ser Leu Gly
        995                1000                1005

cac tat gct gct tta cac gtc gct ggt gtt ctg tcc gcc aat gat acc   3072
His Tyr Ala Ala Leu His Val Ala Gly Val Leu Ser Ala Asn Asp Thr
   1010                1015                1020
```

```
att tac ctc act ggc atc aga gcg cag ctg ctc gtg gat aag tgc cag   3120
Ile Tyr Leu Thr Gly Ile Arg Ala Gln Leu Leu Val Asp Lys Cys Gln
1025                1030                1035                1040

gca gga acc cac tca atg ctg gca gtg agg gca tcc tta cta cag atc   3168
Ala Gly Thr His Ser Met Leu Ala Val Arg Ala Ser Leu Leu Gln Ile
                1045                1050                1055

caa cag ttc ctc gat gcc aac att cac gag gtt gca tgt gtc aat ggt   3216
Gln Gln Phe Leu Asp Ala Asn Ile His Glu Val Ala Cys Val Asn Gly
            1060                1065                1070

tca cga gaa gtc gtc atc agt gga cgc gtt gcc gac att gac cag ctg   3264
Ser Arg Glu Val Val Ile Ser Gly Arg Val Ala Asp Ile Asp Gln Leu
        1075                1080                1085

gtt ggc ctt ttg tcg gct gac aac atc aag gcg acc cgc gtc aag gtg   3312
Val Gly Leu Leu Ser Ala Asp Asn Ile Lys Ala Thr Arg Val Lys Val
    1090                1095                1100

cca ttt gcc ttc cac tca gcg cag gtt gac ccc att ctc tcc gac ttg   3360
Pro Phe Ala Phe His Ser Ala Gln Val Asp Pro Ile Leu Ser Asp Leu
1105                1110                1115                1120

gat aca gcg gcg tcg cgc gtc acc ttc cac tcc ctc cag att cct gtt   3408
Asp Thr Ala Ala Ser Arg Val Thr Phe His Ser Leu Gln Ile Pro Val
                1125                1130                1135

ctt tgt gcc ctt gac agc tct gtc atc agc cct gga aac cat ggt gtc   3456
Leu Cys Ala Leu Asp Ser Ser Val Ile Ser Pro Gly Asn His Gly Val
            1140                1145                1150

att ggt ccc ctt cat cta cag cga cat tgt cgt gag aca gtc aac ttt   3504
Ile Gly Pro Leu His Leu Gln Arg His Cys Arg Glu Thr Val Asn Phe
        1155                1160                1165

gag ggt gct cta cat gct gcg gag cac gag aag atc atc aac aag aca   3552
Glu Gly Ala Leu His Ala Ala Glu His Glu Lys Ile Ile Asn Lys Thr
    1170                1175                1180

tca act cta tgg atc gag att ggt ccc cat gtt gtc tgc tct acc ttc   3600
Ser Thr Leu Trp Ile Glu Ile Gly Pro His Val Val Cys Ser Thr Phe
1185                1190                1195                1200

ctc aag tcc agc ctt ggt cca agc aca cct gct atc gca tcg ctt cgc   3648
Leu Lys Ser Ser Leu Gly Pro Ser Thr Pro Ala Ile Ala Ser Leu Arg
                1205                1210                1215

cga aat gac gat tgc tgg aag gtg ttg gct gat ggt ttg agc agt ctc   3696
Arg Asn Asp Asp Cys Trp Lys Val Leu Ala Asp Gly Leu Ser Ser Leu
            1220                1225                1230

tac agc agc ggg ttg aca att gac ttg aac gag tat cat cgc gac ttc   3744
Tyr Ser Ser Gly Leu Thr Ile Asp Leu Asn Glu Tyr His Arg Asp Phe
        1235                1240                1245
```

```
aag gcc tct cac cag gta ctt cgt ctg cct tgt tac agc tgg gag cac   3792
Lys Ala Ser His Gln Val Leu Arg Leu Pro Cys Tyr Ser Trp Glu His
   1250                1255                1260

aag aat tac tgg ata cag tac aag tac gat tgg tcc ttg gct aaa ggt   3840
Lys Asn Tyr Trp Ile Gln Tyr Lys Tyr Asp Trp Ser Leu Ala Lys Gly
1265                1270                1275                1280

gat cct cca att gcc cct aac agc tcg gtt gaa gca gtc tca gct tta   3888
Asp Pro Pro Ile Ala Pro Asn Ser Ser Val Glu Ala Val Ser Ala Leu
               1285                1290                1295

tca aca ccc tcg gtc cag aag att cta cag gag act tcc ctt gat cag   3936
Ser Thr Pro Ser Val Gln Lys Ile Leu Gln Glu Thr Ser Leu Asp Gln
           1300                1305                1310

gta ttg act atc gtt gct gag aca gat ctc gcg agc cct cta ttg tca   3984
Val Leu Thr Ile Val Ala Glu Thr Asp Leu Ala Ser Pro Leu Leu Ser
       1315                1320                1325

gag gtt gcc caa ggt cat cgg gtc aac ggt gtc aaa gtc tgc aca tct   4032
Glu Val Ala Gln Gly His Arg Val Asn Gly Val Lys Val Cys Thr Ser
   1330                1335                1340

tcc gtg tac gct gat gtt ggc ttg acg ctg ggt aag tac att ttg gac   4080
Ser Val Tyr Ala Asp Val Gly Leu Thr Leu Gly Lys Tyr Ile Leu Asp
1345                1350                1355                1360

aac tac cgc acc gac tta gag ggt tat gcg gtc gat gtt cac ggt att   4128
Asn Tyr Arg Thr Asp Leu Glu Gly Tyr Ala Val Asp Val His Gly Ile
               1365                1370                1375

gag gtc cac aag cca ctt ctt ctc aaa gaa gac atg aac gga acg ccc   4176
Glu Val His Lys Pro Leu Leu Leu Lys Glu Asp Met Asn Gly Thr Pro
           1380                1385                1390

cag gct aca ccg ttc cgc atc gaa gtg cga tac cca atc cag agc acc   4224
Gln Ala Thr Pro Phe Arg Ile Glu Val Arg Tyr Pro Ile Gln Ser Thr
       1395                1400                1405

acg gcg ctg atg agc atc tcc acc act ggc ccc aac ggt cag cac atc   4272
Thr Ala Leu Met Ser Ile Ser Thr Thr Gly Pro Asn Gly Gln His Ile
   1410                1415                1420

aag cat gct aac tgc gaa ctt cga ctc gag cat ccg tcg caa tgg gaa   4320
Lys His Ala Asn Cys Glu Leu Arg Leu Glu His Pro Ser Gln Trp Glu
1425                1430                1435                1440

gcg gag tgg gat cgc caa gcc tac ctc atc aat cgc agc gtc aac tgc   4368
Ala Glu Trp Asp Arg Gln Ala Tyr Leu Ile Asn Arg Ser Val Asn Cys
               1445                1450                1455

ctt ctg cag cga tca gca caa ggt ttg gac agc atg ttg gca acc gga   4416
Leu Leu Gln Arg Ser Ala Gln Gly Leu Asp Ser Met Leu Ala Thr Gly
           1460                1465                1470
```

```
atg gtc tac aag gtc ttc tcc tcc ctc gtc gac tat gcc gat ggc tac   4464
Met Val Tyr Lys Val Phe Ser Ser Leu Val Asp Tyr Ala Asp Gly Tyr
       1475                1480                1485

aag ggt ctg cag gag gtt gtc ttg cac agc caa gag ctc gag ggc aca   4512
Lys Gly Leu Gln Glu Val Val Leu His Ser Gln Glu Leu Glu Gly Thr
   1490                1495                1500

gca aaa gtg cgc ttc caa act ccc tcg gga ggt ttc gtc tgc aat ccc   4560
Ala Lys Val Arg Phe Gln Thr Pro Ser Gly Gly Phe Val Cys Asn Pro
1505                1510                1515                1520

atg tgg att gac agc tgt ggt cag acg acc ggc ttc atg atg aac tgt   4608
Met Trp Ile Asp Ser Cys Gly Gln Thr Thr Gly Phe Met Met Asn Cys
               1525                1530                1535

cat cag act acg ccc aat gac tac gtc tac gtc aat cat ggc tgg aag   4656
His Gln Thr Thr Pro Asn Asp Tyr Val Tyr Val Asn His Gly Trp Lys
           1540                1545                1550

tcg atg aga ttg gcc aag gcg ttc cgt gaa gat ggt acc tat cgg act   4704
Ser Met Arg Leu Ala Lys Ala Phe Arg Glu Asp Gly Thr Tyr Arg Thr
       1555                1560                1565

tat atc cgg atg agg ccc att gat agc acc aag ttc gct ggt gac ttg   4752
Tyr Ile Arg Met Arg Pro Ile Asp Ser Thr Lys Phe Ala Gly Asp Leu
   1570                1575                1580

tac att ctt gat gag gat gac act gtg gtt ggt gtt tat gga gac ata   4800
Tyr Ile Leu Asp Glu Asp Asp Thr Val Val Gly Val Tyr Gly Asp Ile
1585                1590                1595                1600

aca ttc caa ggt ttg ccg cga cga gtt ctc aac aca gtc ttg cca tct   4848
Thr Phe Gln Gly Leu Pro Arg Arg Val Leu Asn Thr Val Leu Pro Ser
               1605                1610                1615

gcc aac gcg gtt cca gtt gat gct ccc atg ggt cga cgg gac gtg cct   4896
Ala Asn Ala Val Pro Val Asp Ala Pro Met Gly Arg Arg Asp Val Pro
           1620                1625                1630

cca tca aga atg gat gtg cct ccc gtc agg tcc ggt gaa ggg cca ccc   4944
Pro Ser Arg Met Asp Val Pro Pro Val Arg Ser Gly Glu Gly Pro Pro
       1635                1640                1645

act tca gca ccc acg cag caa gct atc gct ctg ccg ttc gca gcc gat   4992
Thr Ser Ala Pro Thr Gln Gln Ala Ile Ala Leu Pro Phe Ala Ala Asp
   1650                1655                1660

aca tcc atg gac tcc cga ttg aga cct ctt ctt cgc atc ttg tca gaa   5040
Thr Ser Met Asp Ser Arg Leu Arg Pro Leu Leu Arg Ile Leu Ser Glu
1665                1670                1675                1680

gag atc ggt ctc ggt ctt gac gtt ctt tcg gac gat gaa ctc gac ttt   5088
Glu Ile Gly Leu Gly Leu Asp Val Leu Ser Asp Asp Glu Leu Asp Phe
               1685                1690                1695
```

```
gcg gac cac ggt gtc gac tca ctc ctc tca ttg acc atc act ggt cgc   5136
Ala Asp His Gly Val Asp Ser Leu Leu Ser Leu Thr Ile Thr Gly Arg
          1700                1705                1710

atg cgt gag gaa ttg ggt ctc gac gtt gaa tct aca gca ttc atg aac   5184
Met Arg Glu Glu Leu Gly Leu Asp Val Glu Ser Thr Ala Phe Met Asn
       1715                1720                1725

tgt ccc act ttg ggc agc ttt aaa ttg ttc cta gga ctt gtc gat cag   5232
Cys Pro Thr Leu Gly Ser Phe Lys Leu Phe Leu Gly Leu Val Asp Gln
   1730                1735                1740

gac aat aag ggc agc agc ggc agt gat ggc agt ggt agg agc agt cca   5280
Asp Asn Lys Gly Ser Ser Gly Ser Asp Gly Ser Gly Arg Ser Ser Pro
1745                1750                1755                1760

gca ccg ggt acc gag tct ggc gct act aca cca cct atg agc gaa gag   5328
Ala Pro Gly Thr Glu Ser Gly Ala Thr Thr Pro Pro Met Ser Glu Glu
               1765                1770                1775

gac cag gac aag ata gtc agc agt cac tcg ctt cac cag ttc caa gcc   5376
Asp Gln Asp Lys Ile Val Ser Ser His Ser Leu His Gln Phe Gln Ala
          1780                1785                1790

agt tcg acg ctt cta cag ggc agt ccc agt aaa gct cgc tcg act ttg   5424
Ser Ser Thr Leu Leu Gln Gly Ser Pro Ser Lys Ala Arg Ser Thr Leu
       1795                1800                1805

ttc ttg cta cca gat ggc tcg gga tct gcc aca tcc tac gct tcc ctt   5472
Phe Leu Leu Pro Asp Gly Ser Gly Ser Ala Thr Ser Tyr Ala Ser Leu
   1810                1815                1820

ccc ccg atc tct cca gac gga gat gtt gct gtc tac ggg ttg aac tgt   5520
Pro Pro Ile Ser Pro Asp Gly Asp Val Ala Val Tyr Gly Leu Asn Cys
1825                1830                1835                1840

cca tgg ctg aag gac tct agt tac ctc gtc gag ttt gga ctc aag ggc   5568
Pro Trp Leu Lys Asp Ser Ser Tyr Leu Val Glu Phe Gly Leu Lys Gly
               1845                1850                1855

ttg aca gag ctc tat gtc aac gag ata ctc cgt cgc aag cca cag ggt   5616
Leu Thr Glu Leu Tyr Val Asn Glu Ile Leu Arg Arg Lys Pro Gln Gly
          1860                1865                1870

cct tac aat ttg gga gga tgg tca gcc ggt ggc att tgc gct tat gaa   5664
Pro Tyr Asn Leu Gly Gly Trp Ser Ala Gly Gly Ile Cys Ala Tyr Glu
       1875                1880                1885

gct gcc ctg atc ctc acc aga gca gga cac caa gtc gat cgc ctt atc   5712
Ala Ala Leu Ile Leu Thr Arg Ala Gly His Gln Val Asp Arg Leu Ile
   1890                1895                1900

ttg att gac tct ccc aat ccc gtt ggt ctt gag aag cta cct cct cgc   5760
Leu Ile Asp Ser Pro Asn Pro Val Gly Leu Glu Lys Leu Pro Pro Arg
1905                1910                1915                1920
```

```
ttg tac gat ttc ctc aat tcg cag aat gtc ttt gga tca gac aac ccg   5808
Leu Tyr Asp Phe Leu Asn Ser Gln Asn Val Phe Gly Ser Asp Asn Pro
                1925                1930                1935

cac agc act gct gga aca agc gtc aaa gct cca gaa tgg ctt ctt gca   5856
His Ser Thr Ala Gly Thr Ser Val Lys Ala Pro Glu Trp Leu Leu Ala
            1940                1945                1950

cat ttc ctg gcc ttc att gac gct ctg gat gct tat gtc gca gtg cct   5904
His Phe Leu Ala Phe Ile Asp Ala Leu Asp Ala Tyr Val Ala Val Pro
        1955                1960                1965

tgg gac tct ggt cta gtc ggt cta gca tca ccg ctc cct gca ccg ccg   5952
Trp Asp Ser Gly Leu Val Gly Leu Ala Ser Pro Leu Pro Ala Pro Pro
    1970                1975                1980

cag aca tac atg ctg tgg gca gaa gac gga gtt tgc aaa gac tct gat   6000
Gln Thr Tyr Met Leu Trp Ala Glu Asp Gly Val Cys Lys Asp Ser Asp
1985                1990                1995                2000

agt gct cgt ccc gag tac cgt gac gat gac cca cgc gag atg aga tgg   6048
Ser Ala Arg Pro Glu Tyr Arg Asp Asp Asp Pro Arg Glu Met Arg Trp
                2005                2010                2015

ctg ttg gag aac aga aca aac ttt ggt ccg aat ggt tgg gag gcg cta   6096
Leu Leu Glu Asn Arg Thr Asn Phe Gly Pro Asn Gly Trp Glu Ala Leu
            2020                2025                2030

ctt ggt ggt aaa gag ggt ttg ttc atg gat cgg att gcg gaa gcg aat   6144
Leu Gly Gly Lys Glu Gly Leu Phe Met Asp Arg Ile Ala Glu Ala Asn
        2035                2040                2045

cat ttt agt atg ttg aag aga gga cgg aat gcg gaa tat gtc tct gca   6192
His Phe Ser Met Leu Lys Arg Gly Arg Asn Ala Glu Tyr Val Ser Ala
    2050                2055                2060

ttc ctg gct cgg gcc ttg gac aat tag                              6219
Phe Leu Ala Arg Ala Leu Asp Asn
2065                2070
```

<210> 6
<211> 2072
<212> PRT
<213> Fusarium graminearum

<400> 6

```
Met Thr Pro Ser Met Met Glu Val Phe Val Phe Gly Asp Gln Ser Thr
  1               5                  10                  15

Arg Phe Ala Pro Pro Leu Lys Asp Leu Leu Leu Lys Gly Asn Ser Pro
            20                  25                  30

Tyr Leu Thr His Phe Val Lys Gln Val His Ala Leu Leu Arg Arg Glu
        35                  40                  45
```

Ile Ser Ser Leu Pro Ala Val Gln Gln Lys Leu Phe Pro Asn Phe Ala
50 55 60

Asp Ile Gln Glu Leu Val Ser Lys Ser Asp Trp Gly Ser Gly Asn Pro
65 70 75 80

Ala Leu Thr Ser Ala Leu Ala Cys Phe Tyr His Leu Cys Ser Phe Ile
85 90 95

His Phe Tyr Asp Gly Gln Gly Arg Thr Phe Pro Ser Glu Asn Ser Arg
100 105 110

Ile Ile Gly Leu Cys Val Gly Ser Leu Ala Ala Thr Ala Val Ser Cys
115 120 125

Ser Thr Ser Leu Ser Glu Leu Val Ser Ala Gly Val Asp Ala Val Arg
130 135 140

Val Ala Leu His Val Gly Leu Arg Val Trp Arg Thr Thr Ser Leu Phe
145 150 155 160

Asp Val Pro Asp Arg Pro Ser Ala Thr Trp Phe Ile Ile Val Pro Glu
165 170 175

Ala Val Leu Pro Arg Glu Ser Ala Gln Asp Arg Leu Asp Ser Phe Ile
180 185 190

Ile Glu Met Gly Leu Ala Arg Ser Ser Val Pro Tyr Ile Ser Ser Val
195 200 205

Ala His His Asn Met Thr Ile Ser Gly Pro Pro Ser Val Leu Glu Lys
210 215 220

Phe Ile His Ser Ile Ser Thr Ser Pro Lys Asp Ser Leu Pro Val Pro
225 230 235 240

Ile Tyr Ala Pro Tyr His Ala Ser His Leu Tyr Ser Met Asp Asp Val
245 250 255

Asp Glu Val Leu Ser Leu Ser Ala Pro Ser Phe Ala Ser Glu Ser Ile
260 265 270

Ile Pro Leu Ile Ser Ser Ser Ser Gly Asp Glu Leu Gln Pro Leu Lys
275 280 285

Tyr Ala Asp Leu Leu Arg Cys Cys Val Ser Asp Met Leu Ile Gln Pro
290 295 300

Leu Asp Leu Thr Lys Val Ser Gln Ala Val Ala Gln Leu Leu Glu Val
305 310 315 320

Ser Ser Ser Thr Arg Ala Ile Ile Lys Pro Ile Ala Thr Ser Val Ser
325 330 335

Asn Ser Leu Val Ser Val Leu Glu Pro Thr Leu Ala Glu Arg Cys Ala
340 345 350

```
Val Asp Asn Ser Met Gly Pro Lys Ala Ser Thr Ser His Ser Ser Ala
        355                 360                 365

Glu Thr Gln Thr Glu Ser Ser Ser Lys Asn Ser Lys Ile Ala Ile Val
    370                 375                 380

Ala Met Ser Gly Arg Phe Pro Asp Ala Ala Asp Leu Ser Glu Phe Trp
385                 390                 395                 400

Asp Leu Leu Tyr Glu Gly Arg Asp Val His Arg Gln Ile Pro Glu Asp
                405                 410                 415

Arg Phe Asn Ala Glu Leu His Tyr Asp Ala Thr Gly Arg Arg Lys Asn
            420                 425                 430

Thr Ser Lys Val Met Asn Gly Cys Phe Ile Lys Glu Pro Gly Leu Phe
        435                 440                 445

Asp Ala Arg Phe Phe Asn Met Ser Pro Lys Glu Ala Glu Gln Ser Asp
    450                 455                 460

Pro Gly Gln Arg Met Ala Leu Glu Thr Ala Tyr Glu Ala Leu Glu Met
465                 470                 475                 480

Ala Ser Ile Val Pro Asp Arg Thr Pro Ser Thr Gln Arg Asp Arg Val
                485                 490                 495

Gly Val Phe Tyr Gly Met Thr Ser Asp Asp Trp Arg Glu Val Asn Ser
            500                 505                 510

Gly Gln Asn Val Asp Thr Tyr Phe Ile Pro Gly Gly Asn Arg Ala Phe
        515                 520                 525

Thr Pro Gly Arg Leu Asn Tyr Phe Phe Lys Phe Ser Gly Pro Ser Ala
    530                 535                 540

Ser Val Asp Thr Ala Cys Ser Ser Ser Leu Val Gly Leu His Leu Ala
545                 550                 555                 560

Cys Asn Ser Leu Trp Arg Asn Asp Cys Asp Thr Ala Ile Ala Gly Gly
                565                 570                 575

Thr Asn Val Met Thr Asn Pro Asp Asn Phe Ala Gly Leu Asp Arg Gly
            580                 585                 590

His Phe Leu Ser Arg Thr Gly Asn Cys Asn Thr Phe Asp Asp Gly Ala
        595                 600                 605

Asp Gly Tyr Cys Arg Ala Asp Gly Val Gly Thr Ile Ile Leu Lys Arg
    610                 615                 620

Leu Glu Asp Ala Glu Ala Asp Asn Asp Pro Ile Leu Gly Val Ile Leu
625                 630                 635                 640

Gly Ala Tyr Thr Asn His Ser Ala Glu Ala Val Ser Ile Thr Arg Pro
                645                 650                 655
```

His Ala Gly Ala Gln Glu Tyr Ile Phe Ser Lys Leu Leu Arg Glu Ser
660 665 670

Gly Thr Asp Pro Tyr Asn Val Ser Tyr Ile Glu Met His Gly Thr Gly
675 680 685

Thr Gln Ala Gly Asp Ala Thr Glu Met Thr Ser Val Leu Lys Thr Phe
690 695 700

Ala Pro Thr Ser Gly Phe Gly Gly Arg Leu Pro His Gln Asn Leu His
705 710 715 720

Leu Gly Ser Val Lys Ala Asn Val Gly His Gly Glu Ser Ala Ser Gly
725 730 735

Ile Ile Ala Leu Ile Lys Thr Leu Leu Met Met Glu Lys Asn Met Ile
740 745 750

Pro Pro His Cys Gly Ile Lys Thr Lys Ile Asn His His Phe Pro Thr
755 760 765

Asp Leu Thr Gln Arg Asn Val His Ile Ala Lys Val Pro Thr Ser Trp
770 775 780

Thr Arg Ser Gly Gln Ala Asn Pro Arg Ile Ala Phe Val Asn Asn Phe
785 790 795 800

Ser Ala Ala Gly Gly Asn Ser Ala Val Leu Leu Gln Asp Ala Pro Gln
805 810 815

Pro Ser Val Val Ser Asp Val Thr Asp Pro Arg Thr Ser His Val Val
820 825 830

Thr Met Ser Ala Arg Ser Ala Asp Ser Leu Arg Lys Asn Leu Ala Asn
835 840 845

Leu Lys Glu Leu Val Glu Gly Gln Gly Asp Ser Glu Val Gly Phe Leu
850 855 860

Ser Lys Leu Ser Tyr Thr Thr Thr Ala Arg Arg Met His His Gln Phe
865 870 875 880

Arg Ala Ser Val Thr Ala Gln Thr Arg Glu Gln Leu Leu Lys Gly Leu
885 890 895

Asp Ser Ala Ile Glu Arg Gln Asp Val Lys Arg Ile Pro Ala Ala Ala
900 905 910

Pro Ser Val Gly Phe Val Phe Ser Gly Gln Gly Ala Gln Tyr Arg Gly
915 920 925

Met Gly Lys Glu Tyr Phe Thr Ser Phe Thr Ala Phe Arg Ser Glu Ile
930 935 940

Met Ser Tyr Asp Ser Ile Ala Gln Ala Gln Gly Phe Pro Ser Ile Leu
945 950 955 960

Pro Leu Ile Arg Gly Glu Val Glu Ala Asp Ser Leu Ser Pro Val Glu
965 970 975

Ile Gln Leu Gly Leu Thr Cys Leu Gln Met Ala Leu Ala Lys Leu Trp
980 985 990

Lys Ser Phe Gly Val Glu Pro Gly Phe Val Leu Gly His Ser Leu Gly
995 1000 1005

His Tyr Ala Ala Leu His Val Ala Gly Val Leu Ser Ala Asn Asp Thr
1010 1015 1020

Ile Tyr Leu Thr Gly Ile Arg Ala Gln Leu Leu Val Asp Lys Cys Gln
025 1030 1035 1040

Ala Gly Thr His Ser Met Leu Ala Val Arg Ala Ser Leu Leu Gln Ile
1045 1050 1055

Gln Gln Phe Leu Asp Ala Asn Ile His Glu Val Ala Cys Val Asn Gly
1060 1065 1070

Ser Arg Glu Val Val Ile Ser Gly Arg Val Ala Asp Ile Asp Gln Leu
1075 1080 1085

Val Gly Leu Leu Ser Ala Asp Asn Ile Lys Ala Thr Arg Val Lys Val
1090 1095 1100

Pro Phe Ala Phe His Ser Ala Gln Val Asp Pro Ile Leu Ser Asp Leu
105 1110 1115 1120

Asp Thr Ala Ala Ser Arg Val Thr Phe His Ser Leu Gln Ile Pro Val
1125 1130 1135

Leu Cys Ala Leu Asp Ser Ser Val Ile Ser Pro Gly Asn His Gly Val
1140 1145 1150

Ile Gly Pro Leu His Leu Gln Arg His Cys Arg Glu Thr Val Asn Phe
1155 1160 1165

Glu Gly Ala Leu His Ala Ala Glu His Glu Lys Ile Ile Asn Lys Thr
1170 1175 1180

Ser Thr Leu Trp Ile Glu Ile Gly Pro His Val Val Cys Ser Thr Phe
185 1190 1195 1200

Leu Lys Ser Ser Leu Gly Pro Ser Thr Pro Ala Ile Ala Ser Leu Arg
1205 1210 1215

Arg Asn Asp Asp Cys Trp Lys Val Leu Ala Asp Gly Leu Ser Ser Leu
1220 1225 1230

Tyr Ser Ser Gly Leu Thr Ile Asp Leu Asn Glu Tyr His Arg Asp Phe
1235 1240 1245

Lys Ala Ser His Gln Val Leu Arg Leu Pro Cys Tyr Ser Trp Glu His
1250 1255 1260

Lys Asn Tyr Trp Ile Gln Tyr Lys Tyr Asp Trp Ser Leu Ala Lys Gly
265 1270 1275 1280

Asp Pro Pro Ile Ala Pro Asn Ser Ser Val Glu Ala Val Ser Ala Leu
1285 1290 1295

Ser Thr Pro Ser Val Gln Lys Ile Leu Gln Glu Thr Ser Leu Asp Gln
1300 1305 1310

Val Leu Thr Ile Val Ala Glu Thr Asp Leu Ala Ser Pro Leu Leu Ser
1315 1320 1325

Glu Val Ala Gln Gly His Arg Val Asn Gly Val Lys Val Cys Thr Ser
1330 1335 1340

Ser Val Tyr Ala Asp Val Gly Leu Thr Leu Gly Lys Tyr Ile Leu Asp
345 1350 1355 1360

Asn Tyr Arg Thr Asp Leu Glu Gly Tyr Ala Val Asp Val His Gly Ile
1365 1370 1375

Glu Val His Lys Pro Leu Leu Leu Lys Glu Asp Met Asn Gly Thr Pro
1380 1385 1390

Gln Ala Thr Pro Phe Arg Ile Glu Val Arg Tyr Pro Ile Gln Ser Thr
1395 1400 1405

Thr Ala Leu Met Ser Ile Ser Thr Thr Gly Pro Asn Gly Gln His Ile
1410 1415 1420

Lys His Ala Asn Cys Glu Leu Arg Leu Glu His Pro Ser Gln Trp Glu
425 1430 1435 1440

Ala Glu Trp Asp Arg Gln Ala Tyr Leu Ile Asn Arg Ser Val Asn Cys
1445 1450 1455

Leu Leu Gln Arg Ser Ala Gln Gly Leu Asp Ser Met Leu Ala Thr Gly
1460 1465 1470

Met Val Tyr Lys Val Phe Ser Ser Leu Val Asp Tyr Ala Asp Gly Tyr
1475 1480 1485

Lys Gly Leu Gln Glu Val Val Leu His Ser Gln Glu Leu Glu Gly Thr
1490 1495 1500

Ala Lys Val Arg Phe Gln Thr Pro Ser Gly Gly Phe Val Cys Asn Pro
505 1510 1515 1520

Met Trp Ile Asp Ser Cys Gly Gln Thr Thr Gly Phe Met Met Asn Cys
1525 1530 1535

His Gln Thr Thr Pro Asn Asp Tyr Val Tyr Val Asn His Gly Trp Lys
1540 1545 1550

Ser Met Arg Leu Ala Lys Ala Phe Arg Glu Asp Gly Thr Tyr Arg Thr
1555 1560 1565

Tyr Ile Arg Met Arg Pro Ile Asp Ser Thr Lys Phe Ala Gly Asp Leu
1570 1575 1580

Tyr Ile Leu Asp Glu Asp Asp Thr Val Val Gly Val Tyr Gly Asp Ile
585 1590 1595 1600

Thr Phe Gln Gly Leu Pro Arg Arg Val Leu Asn Thr Val Leu Pro Ser
1605 1610 1615

Ala Asn Ala Val Pro Val Asp Ala Pro Met Gly Arg Arg Asp Val Pro
1620 1625 1630

Pro Ser Arg Met Asp Val Pro Pro Val Arg Ser Gly Glu Gly Pro Pro
1635 1640 1645

Thr Ser Ala Pro Thr Gln Gln Ala Ile Ala Leu Pro Phe Ala Ala Asp
1650 1655 1660

Thr Ser Met Asp Ser Arg Leu Arg Pro Leu Leu Arg Ile Leu Ser Glu
665 1670 1675 1680

Glu Ile Gly Leu Gly Leu Asp Val Leu Ser Asp Asp Glu Leu Asp Phe
1685 1690 1695

Ala Asp His Gly Val Asp Ser Leu Leu Ser Leu Thr Ile Thr Gly Arg
1700 1705 1710

Met Arg Glu Glu Leu Gly Leu Asp Val Glu Ser Thr Ala Phe Met Asn
1715 1720 1725

Cys Pro Thr Leu Gly Ser Phe Lys Leu Phe Leu Gly Leu Val Asp Gln
1730 1735 1740

Asp Asn Lys Gly Ser Ser Gly Ser Asp Gly Ser Gly Arg Ser Ser Pro
745 1750 1755 1760

Ala Pro Gly Thr Glu Ser Gly Ala Thr Thr Pro Pro Met Ser Glu Glu
1765 1770 1775

Asp Gln Asp Lys Ile Val Ser Ser His Ser Leu His Gln Phe Gln Ala
1780 1785 1790

Ser Ser Thr Leu Leu Gln Gly Ser Pro Ser Lys Ala Arg Ser Thr Leu
1795 1800 1805

Phe Leu Leu Pro Asp Gly Ser Gly Ser Ala Thr Ser Tyr Ala Ser Leu
1810 1815 1820

Pro Pro Ile Ser Pro Asp Gly Asp Val Ala Val Tyr Gly Leu Asn Cys
825 1830 1835 1840

Pro Trp Leu Lys Asp Ser Ser Tyr Leu Val Glu Phe Gly Leu Lys Gly
1845 1850 1855

Leu Thr Glu Leu Tyr Val Asn Glu Ile Leu Arg Arg Lys Pro Gln Gly
1860 1865 1870

```
Pro Tyr Asn Leu Gly Gly Trp Ser Ala Gly Gly Ile Cys Ala Tyr Glu
       1875                1880                1885

Ala Ala Leu Ile Leu Thr Arg Ala Gly His Gln Val Asp Arg Leu Ile
   1890                1895                1900

Leu Ile Asp Ser Pro Asn Pro Val Gly Leu Glu Lys Leu Pro Pro Arg
905                1910                1915                1920

Leu Tyr Asp Phe Leu Asn Ser Gln Asn Val Phe Gly Ser Asp Asn Pro
               1925                1930                1935

His Ser Thr Ala Gly Thr Ser Val Lys Ala Pro Glu Trp Leu Leu Ala
           1940                1945                1950

His Phe Leu Ala Phe Ile Asp Ala Leu Asp Ala Tyr Val Ala Val Pro
       1955                1960                1965

Trp Asp Ser Gly Leu Val Gly Leu Ala Ser Pro Leu Pro Ala Pro Pro
   1970                1975                1980

Gln Thr Tyr Met Leu Trp Ala Glu Asp Gly Val Cys Lys Asp Ser Asp
985                1990                1995                2000

Ser Ala Arg Pro Glu Tyr Arg Asp Asp Asp Pro Arg Glu Met Arg Trp
               2005                2010                2015

Leu Leu Glu Asn Arg Thr Asn Phe Gly Pro Asn Gly Trp Glu Ala Leu
           2020                2025                2030

Leu Gly Gly Lys Glu Gly Leu Phe Met Asp Arg Ile Ala Glu Ala Asn
       2035                2040                2045

His Phe Ser Met Leu Lys Arg Gly Arg Asn Ala Glu Tyr Val Ser Ala
   2050                2055                2060

Phe Leu Ala Arg Ala Leu Asp Asn
065                2070
```

<210> 7
<211> 228
<212> DNA
<213> Fusarium graminearum

<220>
<221> CDS
<222> (1)..(228)

<400> 7

```
ggc aac aga gcg ttc act cct ggt cga ctc aac tac ttc ttc aag ttc   48
Gly Asn Arg Ala Phe Thr Pro Gly Arg Leu Asn Tyr Phe Phe Lys Phe
  1               5                  10                  15

agt ggg cct agc gct agt gtt gat acg gct tgc tcc tcc agt ctc gtt   96
Ser Gly Pro Ser Ala Ser Val Asp Thr Ala Cys Ser Ser Ser Leu Val
             20                  25                  30
```

ggc ttg cac ttg gct tgt aat tcc ctc tgg aga aat gat tgc gat aca 144
Gly Leu His Leu Ala Cys Asn Ser Leu Trp Arg Asn Asp Cys Asp Thr
35 40 45

gct att gcg ggc gga acc aat gtc atg act aac cct gac aac ttc gct 192
Ala Ile Ala Gly Gly Thr Asn Val Met Thr Asn Pro Asp Asn Phe Ala
50 55 60

ggt ttg gac cga ggc cac ttc cta tct aga acc ggt 228
Gly Leu Asp Arg Gly His Phe Leu Ser Arg Thr Gly
65 70 75

<210> 8
<211> 76
<212> PRT
<213> Fusarium graminearum

<400> 8
Gly Asn Arg Ala Phe Thr Pro Gly Arg Leu Asn Tyr Phe Phe Lys Phe
1 5 10 15

Ser Gly Pro Ser Ala Ser Val Asp Thr Ala Cys Ser Ser Ser Leu Val
20 25 30

Gly Leu His Leu Ala Cys Asn Ser Leu Trp Arg Asn Asp Cys Asp Thr
35 40 45

Ala Ile Ala Gly Gly Thr Asn Val Met Thr Asn Pro Asp Asn Phe Ala
50 55 60

Gly Leu Asp Arg Gly His Phe Leu Ser Arg Thr Gly
65 70 75

<210> 9
<211> 6534
<212> DNA
<213> Wagiella dermatidis

<400> 9
atggaggagg tctacgtctt cggcgaccaa acggctgact gccgggcgtt cttcaccaag 60
gtctttaccc ggaaggacaa tgtcctcttg cagagcttcc tggaaagagc tggtgaagcc 120
gtcagattcg aaaaccagaa ccgatcacat ccctccaaag cagtccctaa tttctctacc 180
atccaagagc ttgtcgaccg ttattacaga ggtgacgcga aggatgccgc gaccgagagt 240
gctctcgttt gcatttcaca gttttgtcat ttcattggtg cctttgagga gagacggcct 300
tcgtatattc aacctaattc agatgctcga ctagtcggtc tgtgcacagg tctaattgct 360
gcgaccgcag tcgctgcctc agattccctg acagccctta ttcctttggc ggtcgaagcg 420
gtccggatag cgttcagagc tggtgcccac gtgggaaaag ttgcacagca gacagaatgt 480
gactccaaga ctcaatcgtg gtccaccatc gttgctgccg acgagaaatc tgcgcaggag 540
gctctggatg cctttcacaa agagaanggt acatcaccca tcaaccagtt gtggatcagt 600
gtttcttccg ccacttcagt caccattagc gttccgccat ggaccaaagc ccgtcttttn 660
gaagagtcag agttcttccg cacacaaaag agtgctccag tgtcaatatt cgctccntat 720
cacgcatcgc acntacactc gcaatccgat ttggacaaaa tcttacgacc gcagaccaag 780
acaatctttg gcaataccac agtgcggttt cccgtctgct caagcgtgac cggcaaaccg 840

ttcaacgctg aaaatgggtt tgagcttctc caggccgcgt tgaaagaaat cataatcgat 900
cccctgcgat gggacaaggt gttgaaatat tgtgcggcag ggaaggcgtc ggaagccaag 960
gtattcgctg ttggaccgac aaacctggcg agcagtgtgg tctccgctct caaagcctca 1020
acaacgaagg tcacgctgga agaccactcc acctggagta cagtgccacc gcagggaacg 1080
cgtcattcaa aacgagaggc tgatatcgct attgtcggct tctctggtcg cttccccgac 1140
gccgcagata atgagctttt ctggcaattg ttggaacgtg gcttggatgt tcaccggccg 1200
gtccctcccg acaggttccc tgtcgagagc cacactgatc caagtggaaa gaagaagaac 1260
acctctcaca caccgtttgg caattttatt gagaagccag gcttgtttga cgcgaggttc 1320
ttcaatatgt caccaagaga agccgcacaa actgacccca tgcagcggct gatgttgacc 1380
accggctatg angctatgga aatggctggc atcgtccctg gcanaacacc ctcnacnaan 1440
catgacagaa ttggcacttt ctacggtcaa acctcanacn attggcgtga ggtcaatgcc 1500
gcccangata ttgatacata tttcatttca ggtggcgtgc gcgccttcgg acctggtcng 1560
atcaactact tttttaagtt ttccggacct antttctcgg tggacatgcn tgctaatcca 1620
gcttggccgc ntatgaatgt cgcaatcaca tctctcagag caaatgagtg cgatactgtg 1680
ttcacgggtg gtgccaatgt gctcacgaac tctgacattt tctctggtct cagcagaggc 1740
cacttcttgt ccaaaaccgg gtcttgcaag acatgggata atgacgctga tggctactgc 1800
cgcggcgatg gtgtttgcac ggtcatcatg aagcgtcttg acgatgccct tgcggatcga 1860
gaccctgttc tgggtgttat ccgcggtatt ggcacgaacc attcagcaga agccgtttct 1920
atcacccatc cctgtgcgga gaatcaggca ttcttgttcg acaaggtgct caaagagtgc 1980
aacgttcact gcaatgacgt caactacgtt gagatgcacg gcacaggcac gcaggcaggc 2040
gacggcatcg agatggaatc tgtctcatct gtttttgccc cacgacagcc tcgaagacgt 2100
cctgaccagc ctctctacgt tggtgcagtc aagtcaaaca tcggtcacgg tgaggccgtt 2160
tctggcgtat ccgctctgat caaggtgctc cttatgctgc aaaagaacaa gatcccgcct 2220
cataccggca tcaaaaaaca gatcaacaag aactttgcac cagatctcaa ggagcgcaac 2280
gtcaatattg ccttccagac aacgcctttt ccacgcccac ctggtggcaa gcgaactgtg 2340
ttcatcaaca actttagtgc agctggtggt aacacagcga tgcttttgca agatggcccg 2400
gaggtcccaa cggagccttc ttctgaccca agatcgaccc atgtggtgac antctcagcg 2460
aaatctttgg ccgcttttaa gaggaccctt gccaaatacg aagcntattt gaatgcgcac 2520
ccgaatgtcg gccttcccga ccttgcatac actgtcacgg cccgccgggc gcattacagc 2580
ttaccgcgcc gcttcccagt tcagtcaatt tctcagttgc aagcttcgtt gagggccatt 2640
caggaccaga cccacaaccc gatccccctt gcctcacctc agattgctat ggcttttaca 2700
gggcaaggtt cacagtacac tgggatgggc cagaaattgt ttgagacatc gaagcaattc 2760
cgacaagaca tcgaagagtt caatgagatc gctcttcggc aggggctccc ttctattatg 2820
cccttgatcg atggatctgt ggaagttcag catctaccgc ccacagtcgt ccaactcggt 2880
atgtgctgta ttcaaatggc tcttactcat ctttggtcga cttggggaat tcagcccagt 2940
gttgttatcg gtcactcctt gggagaatac gctgctctcc aagccgctgg cgttctgtcc 3000
attgccgaca ccatctatct ggttggtaag cgagctcagc tgctggagca gaagtgcaca 3060
gcaggcactc atgcgatgct tgccgtgaga tctcctgttg gtgggctcca ggatgtggtt 3120
gccaatagcc atggcaagat tgaaaattgc ggcatcaacg gcgtgtctga cactgttctt 3180
tccggcacaa tgggcgatat tgataccgtc gctcaaaaac tggcggacgc tggccagaag 3240
tgcaccaagt tgaagttgcc attcgcattt cattcatccc aagtcgatcc cattttggca 3300
gacttcgaaa aattggcctc ttcggtcaat taccacccac cccgtgttcc tgtcatctcg 3360
cccctgttga gcgatgttgt cagtgtcggg ggcgtcttcg atgcattcta cctcagtaga 3420
cactgccgca agacagtcga ctttgtcggt gggttgtccg ctggtatgtc aaccgccacc 3480
attagcgata cttcgctttg gctcgaggtt ggcggtcacc ccctctgcgc cagcatgatc 3540
aaatcttgct tgtcagtccc aaccctggcc acgatgcgcc gtgatgaaga cccctggaag 3600
atcatttctg cgtccatggc cggcctctat acggccggga agtctttgaa ctgggatgca 3660
ttccacaagg agaacgagtc cttgcgcgtg ctcaacgact tgccattcta tggattcgac 3720
gagaagaatt actggttaca gtacactgga gattggcttt tgtacaaggg cgactacccc 3780

aaggcgattg ccccagctcc agccgccgct gcagcagcaa ggcctgccaa agcgaggaaa 3840
tatctgtcaa cctccgttca aggtattgtc tccgaagagg tcaagggcaa gactgtcacg 3900
attgtcgccg agtccgactt tgcgcacccg aaattgttcc cggtcattgc cggtcacctt 3960
gtcaacggca gtggtctctg cccgtccaca ttgtatgccg acatggccta cacccttggt 4020
caactaggcg ttggcctcct gaagccgggt gagaaggtgg acatcaatat tggcacaatg 4080
gacaatccgg cacctcttct gctcaagaac atcaatcagc ctgaaagcca aattgttcaa 4140
atgacaatga agatcgatct cgatgccagg aaagcggact tcgctgtgac cagcaacaat 4200
ggaaagaaag atgtcactca tgccaagtgc gtgatcgtct tcgaagatgc tgctgtttgg 4260
aaagaacaat ggagcaagac ttcttacctg atccagtcga gaatcgacat gttgaaacac 4320
aagatggaga atggtgaagc cgacaaggtt agtcgcgcta tggcttacaa gctcttcggc 4380
gccctcgttg actacgccga tatattccaa ggcatgcaaa acgtggtctt tgatggaccg 4440
gaattcgagg cgacgagcaa catcaaattc cgcgctgggc cgaacgatgg cgacttttac 4500
ttcagcccct atttcattga cagcgcttgc cacctctcan gcttcaccgt cnatgctacc 4560
gtcnacccac aagatgagtg ctacatttct catggttggt ccagtctccg cttcattgag 4620
ccgcttcagc atgaccagca atactacgca tacctcaaga tgcagcccgt ttcaggaagc 4680
aagatnagag ctggtgatgt ttatgtcttc aacgcggata aagangtcct ccgtcttgcg 4740
ggtggtgtgc gattccagtg cattccccgc aagctcatgg atgtgatgat gcccaaacct 4800
aaggccaatg caaaggctgc ttccccagca tctgcgaccc cgaaagcagc tccaactgtc 4860
aaagcctcca gcaacctcgc tgctcctgct tccgtggaac tgcccactcc caaggtcaag 4920
gcgaagaagc cggttaaggc acaaaagatc aaggcaccag caccacccaa gcaagcctct 4980
tctggtagtc ttgtcgtccg ggctttcgaa atcatcgcaa aggagattga tgtcgaccaa 5040
tctgaactca atgacgatat ccaatgggct gacatgggtg ttgacagctt gatgtcgctg 5100
accatctctg gcaagttcag ggaagatctt gacctcgagg ttgngagcac cctcttcacc 5160
gattacgcct ccgtgggcgc ccttcggaag catctcagcg gcatgtctgg tcctgagccc 5220
attgctactg gtgatgcaag ctcggttgaa tctacggact cgggttcgga gagcgacgag 5280
gataccgtcg agtcgggcat caccaccccg gacaccgaag anttcccagc gaagcntcag 5340
gaacagggga aatccgctgc tgtggaggcg atggctcaac caccttccgc cgaagggcag 5400
gatatgattg agaccattcg ngtagtcatc gcccaagaaa tggagatgga tctggctgaa 5460
atcacggacg ntactgacct cagtaacctc ggaatggact ccttgatggc cctgaccgtt 5520
cttgggaagc ttcgagagga tcatgacatt gatttggatc cgaccatctt ggcggacaat 5580
ccaaccctgg cgcacctccg caaagcattg ggtcttgaga aagccaagcc ggcgcctgca 5640
ccgaaacaag angtcaggac caatgtcgtt gtcgctcctg ctgcgcctcc ggttgangtt 5700
gtcgtgcana tnccccccgc cacatctgtg ttgcttcagg gcaatcccaa gacagccacc 5760
aanaatctgt tcttgttccc tgatggctcc gggtctgcca catcctatgt gtctatcccg 5820
gccattgaca gcaanaacct ggctgtctat gggttaaatt gtccattcat gaaagatccc 5880
accagctata cttgcgggat cnaaagtgtc tccaanctgt atcttgaaaa agtccttcnc 5940
cgacagccta atggtccata tatcctcngg ggctggtcag caagtggtgt ctttgcttac 6000
naaatcacca nacaactcan tgacttacag aanctccacc ctgacaagaa ctacaccgtt 6060
gaaaaattga atcttatcna ctcnccatgc ccgatccgtc tggagcctct acctgcccgc 6120
ctgcaccact tctttgatga aattggtctt ttggggacag gtaccggcaa gaccccgaac 6180
tggctgctac cccatttcga atacagcatc aaggcgttga ctgcctaccg ccctgagctg 6240
aagtctacac gtgatttcaa tgcaccaccc accttgctca tctgggccac tgatggtgtc 6300
tgcggcaagc ccggtgaccc gagaccgccg ccacaagcgg atgatccgaa gagcatgaag 6360
tggctgctgg aaaaccgcac ggactttgga cccaatggat gggacaagct tttgggtgct 6420
gaagtctgta aaatggttac agtggttggt aaccatttca ccatgatgaa accaccggtg 6480
gctaagggtg ttggtcaata catccgggag tctctctcca tgtanagagc ttga 6534

<210> 10
<211> 2177

<212> PRT
<213> Wagiella dermatidis

<400> 10

```
Met Glu Glu Val Tyr Val Phe Gly Asp Gln Thr Ala Asp Cys Arg Ala
 1               5                  10                  15

Phe Phe Thr Lys Val Phe Thr Arg Lys Asp Asn Val Leu Leu Gln Ser
            20                  25                  30

Phe Leu Glu Arg Ala Gly Glu Ala Val Arg Phe Glu Asn Gln Asn Arg
        35                  40                  45

Ser His Pro Ser Lys Ala Val Pro Asn Phe Ser Thr Ile Gln Glu Leu
    50                  55                  60

Val Asp Arg Tyr Tyr Arg Gly Asp Ala Lys Asp Ala Ala Thr Glu Ser
65                  70                  75                  80

Ala Leu Val Cys Ile Ser Gln Phe Cys His Phe Ile Gly Ala Phe Glu
                85                  90                  95

Glu Arg Arg Pro Ser Tyr Ile Gln Pro Asn Ser Asp Ala Arg Leu Val
            100                 105                 110

Gly Leu Cys Thr Gly Leu Ile Ala Ala Thr Ala Val Ala Ala Ser Asp
        115                 120                 125

Ser Leu Thr Ala Leu Ile Pro Leu Ala Val Glu Ala Val Arg Ile Ala
    130                 135                 140

Phe Arg Ala Gly Ala His Val Gly Lys Val Ala Gln Gln Thr Glu Cys
145                 150                 155                 160

Asp Ser Lys Thr Gln Ser Trp Ser Thr Ile Val Ala Ala Asp Glu Lys
                165                 170                 175

Ser Ala Gln Glu Ala Leu Asp Ala Phe His Lys Glu Xaa Gly Thr Ser
            180                 185                 190

Pro Ile Asn Gln Leu Trp Ile Ser Val Ser Ser Ala Thr Ser Val Thr
        195                 200                 205

Ile Ser Val Pro Pro Trp Thr Lys Ala Arg Leu Xaa Glu Glu Ser Glu
    210                 215                 220

Phe Phe Arg Thr Gln Lys Ser Ala Pro Val Ser Ile Phe Ala Pro Tyr
225                 230                 235                 240

His Ala Ser His Xaa His Ser Gln Ser Asp Leu Asp Lys Ile Leu Arg
                245                 250                 255

Pro Gln Thr Lys Thr Ile Phe Gly Asn Thr Thr Val Arg Phe Pro Val
            260                 265                 270
```

Cys Ser Ser Val Thr Gly Lys Pro Phe Asn Ala Glu Asn Gly Phe Glu
275 280 285

Leu Leu Gln Ala Ala Leu Lys Glu Ile Ile Ile Asp Pro Leu Arg Trp
290 295 300

Asp Lys Val Leu Lys Tyr Cys Ala Ala Gly Lys Ala Ser Glu Ala Lys
305 310 315 320

Val Phe Ala Val Gly Pro Thr Asn Leu Ala Ser Ser Val Val Ser Ala
325 330 335

Leu Lys Ala Ser Thr Thr Lys Val Thr Leu Glu Asp His Ser Thr Trp
340 345 350

Ser Thr Val Pro Pro Gln Gly Thr Arg His Ser Lys Arg Glu Ala Asp
355 360 365

Ile Ala Ile Val Gly Phe Ser Gly Arg Phe Pro Asp Ala Ala Asp Asn
370 375 380

Glu Leu Phe Trp Gln Leu Leu Glu Arg Gly Leu Asp Val His Arg Pro
385 390 395 400

Val Pro Pro Asp Arg Phe Pro Val Glu Ser His Thr Asp Pro Ser Gly
405 410 415

Lys Lys Lys Asn Thr Ser His Thr Pro Phe Gly Asn Phe Ile Glu Lys
420 425 430

Pro Gly Leu Phe Asp Ala Arg Phe Phe Asn Met Ser Pro Arg Glu Ala
435 440 445

Ala Gln Thr Asp Pro Met Gln Arg Leu Met Leu Thr Thr Gly Tyr Xaa
450 455 460

Ala Met Glu Met Ala Gly Ile Val Pro Gly Xaa Thr Pro Ser Thr Xaa
465 470 475 480

His Asp Arg Ile Gly Thr Phe Tyr Gly Gln Thr Ser Xaa Xaa Trp Arg
485 490 495

Glu Val Asn Ala Ala Xaa Asp Ile Asp Thr Tyr Phe Ile Ser Gly Gly
500 505 510

Val Arg Ala Phe Gly Pro Gly Xaa Ile Asn Tyr Phe Phe Lys Phe Ser
515 520 525

Gly Pro Xaa Phe Ser Val Asp Met Xaa Ala Asn Pro Ala Trp Pro Xaa
530 535 540

Met Asn Val Ala Ile Thr Ser Leu Arg Ala Asn Glu Cys Asp Thr Val
545 550 555 560

Phe Thr Gly Gly Ala Asn Val Leu Thr Asn Ser Asp Ile Phe Ser Gly
565 570 575

Leu Ser Arg Gly His Phe Leu Ser Lys Thr Gly Ser Cys Lys Thr Trp
580 585 590

Asp Asn Asp Ala Asp Gly Tyr Cys Arg Gly Asp Gly Val Cys Thr Val
595 600 605

Ile Met Lys Arg Leu Asp Asp Ala Leu Ala Asp Arg Asp Pro Val Leu
610 615 620

Gly Val Ile Arg Gly Ile Gly Thr Asn His Ser Ala Glu Ala Val Ser
625 630 635 640

Ile Thr His Pro Cys Ala Glu Asn Gln Ala Phe Leu Phe Asp Lys Val
645 650 655

Leu Lys Glu Cys Asn Val His Cys Asn Asp Val Asn Tyr Val Glu Met
660 665 670

His Gly Thr Gly Thr Gln Ala Gly Asp Gly Ile Glu Met Glu Ser Val
675 680 685

Ser Ser Val Phe Ala Pro Arg Gln Pro Arg Arg Arg Pro Asp Gln Pro
690 695 700

Leu Tyr Val Gly Ala Val Lys Ser Asn Ile Gly His Gly Glu Ala Val
705 710 715 720

Ser Gly Val Ser Ala Leu Ile Lys Val Leu Leu Met Leu Gln Lys Asn
725 730 735

Lys Ile Pro Pro His Thr Gly Ile Lys Lys Gln Ile Asn Lys Asn Phe
740 745 750

Ala Pro Asp Leu Lys Glu Arg Asn Val Asn Ile Ala Phe Gln Thr Thr
755 760 765

Pro Phe Pro Arg Pro Pro Gly Gly Lys Arg Thr Val Phe Ile Asn Asn
770 775 780

Phe Ser Ala Ala Gly Gly Asn Thr Ala Met Leu Leu Gln Asp Gly Pro
785 790 795 800

Glu Val Pro Thr Glu Pro Ser Ser Asp Pro Arg Ser Thr His Val Val
805 810 815

Thr Xaa Ser Ala Lys Ser Leu Ala Ala Phe Lys Arg Thr Leu Ala Lys
820 825 830

Tyr Glu Ala Tyr Leu Asn Ala His Pro Asn Val Gly Leu Pro Asp Leu
835 840 845

Ala Tyr Thr Val Thr Ala Arg Arg Ala His Tyr Ser Leu Pro Arg Arg
850 855 860

Phe Pro Val Gln Ser Ile Ser Gln Leu Gln Ala Ser Leu Arg Ala Ile
865 870 875 880

Gln Asp Gln Thr His Asn Pro Ile Pro Leu Ala Ser Pro Gln Ile Ala
885 890 895

Met Ala Phe Thr Gly Gln Gly Ser Gln Tyr Thr Gly Met Gly Gln Lys
900 905 910

Leu Phe Glu Thr Ser Lys Gln Phe Arg Gln Asp Ile Glu Glu Phe Asn
915 920 925

Glu Ile Ala Leu Arg Gln Gly Leu Pro Ser Ile Met Pro Leu Ile Asp
930 935 940

Gly Ser Val Glu Val Gln His Leu Pro Pro Thr Val Val Gln Leu Gly
945 950 955 960

Met Cys Cys Ile Gln Met Ala Leu Thr His Leu Trp Ser Thr Trp Gly
965 970 975

Ile Gln Pro Ser Val Val Ile Gly His Ser Leu Gly Glu Tyr Ala Ala
980 985 990

Leu Gln Ala Ala Gly Val Leu Ser Ile Ala Asp Thr Ile Tyr Leu Val
995 1000 1005

Gly Lys Arg Ala Gln Leu Leu Glu Gln Lys Cys Thr Ala Gly Thr His
1010 1015 1020

Ala Met Leu Ala Val Arg Ser Pro Val Gly Gly Leu Gln Asp Val Val
1025 1030 1035 1040

Ala Asn Ser His Gly Lys Ile Glu Asn Cys Gly Ile Asn Gly Val Ser
1045 1050 1055

Asp Thr Val Leu Ser Gly Thr Met Gly Asp Ile Asp Thr Val Ala Gln
1060 1065 1070

Lys Leu Ala Asp Ala Gly Gln Lys Cys Thr Lys Leu Lys Leu Pro Phe
1075 1080 1085

Ala Phe His Ser Ser Gln Val Asp Pro Ile Leu Ala Asp Phe Glu Lys
1090 1095 1100

Leu Ala Ser Ser Val Asn Tyr His Pro Pro Arg Val Pro Val Ile Ser
1105 1110 1115 1120

Pro Leu Leu Ser Asp Val Val Ser Val Gly Gly Val Phe Asp Ala Phe
1125 1130 1135

Tyr Leu Ser Arg His Cys Arg Lys Thr Val Asp Phe Val Gly Gly Leu
1140 1145 1150

Ser Ala Gly Met Ser Thr Ala Thr Ile Ser Asp Thr Ser Leu Trp Leu
1155 1160 1165

Glu Val Gly Gly His Pro Leu Cys Ala Ser Met Ile Lys Ser Cys Leu
1170 1175 1180

Ser Val Pro Thr Leu Ala Thr Met Arg Arg Asp Glu Asp Pro Trp Lys
1185 1190 1195 1200

Ile Ile Ser Ala Ser Met Ala Gly Leu Tyr Thr Ala Gly Lys Ser Leu
1205 1210 1215

Asn Trp Asp Ala Phe His Lys Glu Asn Glu Ser Leu Arg Val Leu Asn
1220 1225 1230

Asp Leu Pro Phe Tyr Gly Phe Asp Glu Lys Asn Tyr Trp Leu Gln Tyr
1235 1240 1245

Thr Gly Asp Trp Leu Leu Tyr Lys Gly Asp Tyr Pro Lys Ala Ile Ala
1250 1255 1260

Pro Ala Pro Ala Ala Ala Ala Ala Ala Arg Pro Ala Lys Ala Arg Lys
1265 1270 1275 1280

Tyr Leu Ser Thr Ser Val Gln Gly Ile Val Ser Glu Glu Val Lys Gly
1285 1290 1295

Lys Thr Val Thr Ile Val Ala Glu Ser Asp Phe Ala His Pro Lys Leu
1300 1305 1310

Phe Pro Val Ile Ala Gly His Leu Val Asn Gly Ser Gly Leu Cys Pro
1315 1320 1325

Ser Thr Leu Tyr Ala Asp Met Ala Tyr Thr Leu Gly Gln Leu Gly Val
1330 1335 1340

Gly Leu Leu Lys Pro Gly Glu Lys Val Asp Ile Asn Ile Gly Thr Met
1345 1350 1355 1360

Asp Asn Pro Ala Pro Leu Leu Leu Lys Asn Ile Asn Gln Pro Glu Ser
1365 1370 1375

Gln Ile Val Gln Met Thr Met Lys Ile Asp Leu Asp Ala Arg Lys Ala
1380 1385 1390

Asp Phe Ala Val Thr Ser Asn Asn Gly Lys Lys Asp Val Thr His Ala
1395 1400 1405

Lys Cys Val Ile Val Phe Glu Asp Ala Ala Val Trp Lys Glu Gln Trp
1410 1415 1420

Ser Lys Thr Ser Tyr Leu Ile Gln Ser Arg Ile Asp Met Leu Lys His
1425 1430 1435 1440

Lys Met Glu Asn Gly Glu Ala Asp Lys Val Ser Arg Ala Met Ala Tyr
1445 1450 1455

Lys Leu Phe Gly Ala Leu Val Asp Tyr Ala Asp Ile Phe Gln Gly Met
1460 1465 1470

Gln Asn Val Val Phe Asp Gly Pro Glu Phe Glu Ala Thr Ser Asn Ile
1475 1480 1485

Lys Phe Arg Ala Gly Pro Asn Asp Gly Asp Phe Tyr Phe Ser Pro Tyr
1490 1495 1500

Phe Ile Asp Ser Ala Cys His Leu Ser Xaa Phe Thr Val Xaa Ala Thr
1505 1510 1515 1520

Val Xaa Pro Gln Asp Glu Cys Tyr Ile Ser His Gly Trp Ser Ser Leu
1525 1530 1535

Arg Phe Ile Glu Pro Leu Gln His Asp Gln Gln Tyr Tyr Ala Tyr Leu
1540 1545 1550

Lys Met Gln Pro Val Ser Gly Ser Lys Xaa Arg Ala Gly Asp Val Tyr
1555 1560 1565

Val Phe Asn Ala Asp Lys Xaa Val Leu Arg Leu Ala Gly Gly Val Arg
1570 1575 1580

Phe Gln Cys Ile Pro Arg Lys Leu Met Asp Val Met Met Pro Lys Pro
1585 1590 1595 1600

Lys Ala Asn Ala Lys Ala Ala Ser Pro Ala Ser Ala Thr Pro Lys Ala
1605 1610 1615

Ala Pro Thr Val Lys Ala Ser Ser Asn Leu Ala Ala Pro Ala Ser Val
1620 1625 1630

Glu Leu Pro Thr Pro Lys Val Lys Ala Lys Lys Pro Val Lys Ala Gln
1635 1640 1645

Lys Ile Lys Ala Pro Ala Pro Pro Lys Gln Ala Ser Ser Gly Ser Leu
1650 1655 1660

Val Val Arg Ala Phe Glu Ile Ile Ala Lys Glu Ile Asp Val Asp Gln
1665 1670 1675 1680

Ser Glu Leu Asn Asp Asp Ile Gln Trp Ala Asp Met Gly Val Asp Ser
1685 1690 1695

Leu Met Ser Leu Thr Ile Ser Gly Lys Phe Arg Glu Asp Leu Asp Leu
1700 1705 1710

Glu Val Xaa Ser Thr Leu Phe Thr Asp Tyr Ala Ser Val Gly Ala Leu
1715 1720 1725

Arg Lys His Leu Ser Gly Met Ser Gly Pro Glu Pro Ile Ala Thr Gly
1730 1735 1740

Asp Ala Ser Ser Val Glu Ser Thr Asp Ser Gly Ser Glu Ser Asp Glu
1745 1750 1755 1760

Asp Thr Val Glu Ser Gly Ile Thr Thr Pro Asp Thr Glu Xaa Phe Pro
1765 1770 1775

Ala Lys Xaa Gln Glu Gln Gly Lys Ser Ala Ala Val Glu Ala Met Ala
1780 1785 1790

Gln Pro Pro Ser Ala Glu Gly Gln Asp Met Ile Glu Thr Ile Arg Val
1795 1800 1805

Val Ile Ala Gln Glu Met Glu Met Asp Leu Ala Glu Ile Thr Asp Xaa
1810 1815 1820

Thr Asp Leu Ser Asn Leu Gly Met Asp Ser Leu Met Ala Leu Thr Val
1825 1830 1835 1840

Leu Gly Lys Leu Arg Glu Asp His Asp Ile Asp Leu Asp Pro Thr Ile
1845 1850 1855

Leu Ala Asp Asn Pro Thr Leu Ala His Leu Arg Lys Ala Leu Gly Leu
1860 1865 1870

Glu Lys Ala Lys Pro Ala Pro Ala Pro Lys Gln Xaa Val Arg Thr Asn
1875 1880 1885

Val Val Val Ala Pro Ala Ala Pro Pro Val Xaa Val Val Val Xaa Xaa
1890 1895 1900

Pro Pro Ala Thr Ser Val Leu Leu Gln Gly Asn Pro Lys Thr Ala Thr
1905 1910 1915 1920

Xaa Asn Leu Phe Leu Phe Pro Asp Gly Ser Gly Ser Ala Thr Ser Tyr
1925 1930 1935

Val Ser Ile Pro Ala Ile Asp Ser Xaa Asn Leu Ala Val Tyr Gly Leu
1940 1945 1950

Asn Cys Pro Phe Met Lys Asp Pro Thr Ser Tyr Thr Cys Gly Ile Xaa
1955 1960 1965

Ser Val Ser Xaa Leu Tyr Leu Glu Lys Val Leu Xaa Arg Gln Pro Asn
1970 1975 1980

Gly Pro Tyr Ile Leu Xaa Gly Trp Ser Ala Ser Gly Val Phe Ala Tyr
1985 1990 1995 2000

Xaa Ile Thr Xaa Gln Leu Xaa Asp Leu Gln Xaa Leu His Pro Asp Lys
2005 2010 2015

Asn Tyr Thr Val Glu Lys Leu Asn Leu Ile Xaa Ser Pro Cys Pro Ile
2020 2025 2030

Arg Leu Glu Pro Leu Pro Ala Arg Leu His His Phe Phe Asp Glu Ile
2035 2040 2045

Gly Leu Leu Gly Thr Gly Thr Gly Lys Thr Pro Asn Trp Leu Leu Pro
2050 2055 2060

His Phe Glu Tyr Ser Ile Lys Ala Leu Thr Ala Tyr Arg Pro Glu Leu
2065 2070 2075 2080

Lys Ser Thr Arg Asp Phe Asn Ala Pro Pro Thr Leu Leu Ile Trp Ala
2085 2090 2095

Thr Asp Gly Val Cys Gly Lys Pro Gly Asp Pro Arg Pro Pro Pro Gln
2100 2105 2110

Ala Asp Asp Pro Lys Ser Met Lys Trp Leu Leu Glu Asn Arg Thr Asp
2115 2120 2125

Phe Gly Pro Asn Gly Trp Asp Lys Leu Leu Gly Ala Glu Val Cys Lys
2130 2135 2140

Met Val Thr Val Val Gly Asn His Phe Thr Met Met Lys Pro Pro Val
2145 2150 2155 2160

Ala Lys Gly Val Gly Gln Tyr Ile Arg Glu Ser Leu Ser Met Xaa Arg
2165 2170 2175

Ala

<210> 11
<211> 6330
<212> DNA
<213> Aspergillus parasiticus

<400> 11
atggctcaat caaggcaact ctttctcttc ggcgatcaga cagcggattt tgttcccaag 60
ctccgcagtt tactatccgt ccaggacagc cctattctag ccgcctttct tgaccagtcc 120
cactatgtcg tgcgagccca gatgctgcag agcatgaaca cggttgatca caagttggct 180
cgaaccgctg acctgcgcca aatggtccag aagtatgtcg acggcaaact gacccctgca 240
tttcgaaccg ctctagtgtg cctctgccag ttgggatgct tcatccggga atatgaggaa 300
tctggcaaca tgtacccaca gcccagtgac agctacgtgc tgggattctg catgggttcc 360
ttggccgctg tggcggtaag ctgcagtcgc tccctgtcag agctgctgcc tatcgctgta 420
caaactgtgt tgattgcctt ccgcctcggt ctttgcgccc tggagatgcg ggatcgggtg 480
gatgggtgta gcgatgatcg aggtgaccct tggtctacca ttgtttgggg tctggatccc 540
cagcaagctc gtgatcagat tgaagtgttc tgtcggacca caaacgtacc tcagacaagg 600
cgtccgtgga tcagctgcat ctctaagaat gccatcaccc taagtggcag tccatccact 660
ttgagggcgt tctgtgcgat gcctcagatg gcccagcacc ggactgcccc aattcccatc 720
tgtttaccgg cccacaatgg cgccctcttc acgcaggcag atatcactac catactagac 780
acgacgccta ccactccttg ggagcaactg cccggccaaa taccttatat ttcccatgtc 840
acggggaatg tagtccagac ttccaactac cgggacctta tagaggtagc cctgtctgag 900
actctcttgg agcaagtgcg actagacttg gttgagactg gactgccacg ccttttgcaa 960
tctcgtcagg tcaagagcgt caccatcgta ccattcttga ctcgcatgaa tgagacaatg 1020
agcaacattc tcccagacag ctttatcagt acagagacaa ggactgacac cggacgagcc 1080
atcccagctt caggtcgacc aggcgcaggc aagtgcaagc tggctattgt gtccatgtcg 1140
gggaggttcc ctgaatcacc gaccaccgaa agcttttggg accttctata caaagggttg 1200
gatgtttgta aagaggttcc ccgtcgacgg tgggacatca acacgcatgt ggatcccagc 1260
gggaaagcac gaaacaaagg ggctaccaaa tggggctgct ggctagattt ctcaggcgat 1320
tttgatcccc gattctttgg gatctcgccc aaagaggcgc cacagatgga tccagctcag 1380
cgcatggcct tgatgtctac ttacgaggca atggagcggg ctggtttggt tcccgacacc 1440
acgccgtcga cccagcgaga ccgcattggg gtcttccacg gagtcaccag taacgactgg 1500
atggagacca atacagccca gaacattgac acatacttca tcaccggtgg aaatcggggg 1560
tttattcccg ggcgcattaa cttctgtttc gaatttgccg gacccagcta taccaatgac 1620
acggcctgtt catccagtct agctgccatc cacctggcct gcaattctct ctggcggggc 1680

gactgtgaca cggcggtggc aggaggaact aacatgatct atactcctga tggtcacaca 1740
ggattggaca aagggttctt tctttcccgg actggcaact gcaaacccta cgacgacaag 1800
gccgatggtt actgccgagc tgaggggggtc gggacggtgt tcatcaaacg gctggaagat 1860
gctctggcag ataatgaccc catccttggc gttattctag atgctaaaac taatcactca 1920
gccatgtcgg agtcgatgac tcggccgcac gtgggcgccc aaatcgataa catgacggcg 1980
gcgctgaata ccactggact ccatcccaat gactttagct acattgagat gcatggcact 2040
ggcacccagg taggggatgc ggtggagatg gagtcagtcc tgtcggtgtt tgcgccgtcc 2100
gaaaccgcca gaaaggcgga tcagccacta tttgtcggct cagccaaggc caacgtagga 2160
catggagagg gagtgtctgg ggttacgagc cttattaagg ttctgatgat gatgcagcac 2220
gataccatac ctcctcactg cggcatcaaa ccgggcagca aaatcaaccg caacttccct 2280
gatcttggag ctcgcaatgt gcacatcgcc tttgaaccca agccctggcc acgaacacac 2340
actccgcgca gggtgcttat caacaacttc agtgccgcgg gagggaatac tgccttgata 2400
gtggaagacg ctccggagcg tcactggccg acagagaagg atccgcgctc tagtcatatc 2460
gtcgccctgt ctgcgcatgt gggggcttcc atgaaaacca acctcgaacg actgcatcag 2520
tatctcctga aaaaccccca cactgatctc gcgcagctgt catatactac tactgcgcgt 2580
cgatggcatt atctacaccg agtgagcgtc actggcgcgt ctgttgaaga agtgactcgc 2640
aagctagaga tggccataca gaacggggac ggagtcagtc gacccaaaag caagccgaag 2700
attctctttg ctttcacggg acaagggtct caatatgcaa ctatgggtaa gcaggtgtac 2760
gatgcgtatc catctttcag agaggacctg gagaagtttg atcggttggc gcaaagtcat 2820
ggcttcccta gctttcttca cgtctgtact tcacctaaag gggatgtgga agagatggct 2880
cccgttgtgg tgcaactggc tatcacttgt ctccaaatgg cccttactaa cctcatgacc 2940
tccttcggga tccgtcccga tgtgacagtg gggcatagtt tgggtgaatt tgcagccctg 3000
tatgcggcgg gagttctttc ggcctcagac gtcgtttacc ttgttggtca aagagcggag 3060
ctactccagg agcgctgcca acgcgggacg catgccatgc tggctgtgaa agctacccct 3120
gaagcgttgt cccaatggat ccaggatcat gactgtgagg tggcctgtat taatggccct 3180
gaagataccg ttctcagtgg caccactaag aatgttgccg aggttcaacg cgctatgacg 3240
gacaacggga tcaaatgcac gctgttgaaa ctgccgtttg ccttccattc tgcccaggtg 3300
caacctattc tggacgactt tgaggccctg gctcagggag cgacatttgc caagcctcaa 3360
ctactaattc tctctccctt gctgcggaca gaaatccacg aacaaggcgt cgtgactcca 3420
tcatatgtcg cgcaacattg tcgtcacacc gtagatatgg cccaagcttt gagatctgct 3480
cgagaaaagg gactcatcga cgacaaaacc ctcgtcattg agctgggacc gaagccatta 3540
atctcgggca tggtgaaaat gacactggga gacaaaatta gcaccttacc cactctagca 3600
cctaacaagg ccatttggcc cagcctgcag aagattctca cctcggtcta cacgggtggg 3660
tgggatatta attggaagaa atatcacgcc cctttcgcct cctcccagaa ggtggtggat 3720
ctgccgagct acggctggga tttgaaggac tactacatcc cgtatcaggg tgactggtgt 3780
ctgcatcgcc accagcagga ttgtaagtgc gccgctcctg gccacgaaat caaaacggcc 3840
gactaccaag tgcctcctga gtcgacgcct caccgtccat ccaagctgga ccctagcaag 3900
gaggccttcc ccgaaatcaa gaccaccacg acactccatc gagtggtgga agagacgacc 3960
aaacctttgg gcgccaccct agttgtggag acagacatat ctcggaagga tgtcaacggc 4020
ctcgctcgag ggcaccttgt cgatgggatc cctttgtgta ccccttcctt ttatgctgac 4080
atcgccatgc aagtgggcca atacagtatg caacggctcc gtgcgggaca tccgggggcc 4140
ggtgccatag atggccttgt ggacgtgtcc gacatggtgg tggacaaagc gttggttccc 4200
catgggaagg gacctcaatt gctgcgcacg acgcttacca tggagtggcc gcccaaggct 4260
gctgctacta cgcgaagcgc caaagtcaaa ttcgccacct attttgccga tgggaagctc 4320
gatacggagc atgccagctg tactgtcaga ttcacaagcg atgcacagtt gaaatctcta 4380
cgccggtctg tgtccgagta caagacccac attcgtcagt tacatgatgg ccatgctaag 4440
ggacagttca tgcgatacaa taggaagacc gggtacaagc tcatgagcag catggctcgg 4500
tttaatcccg actacatgct cctagattat ctggtgctga acgaagcaga gaacgaggca 4560
gcaagtggtg tagacttctc gttgggatcg tcggaaggca ccttcgcagc tcacccagct 4620

cacgtcgatg ccatcactca ggtggccggc tttgctatga atgccaatga caatgtcgac 4680
attgagaaac aggtctacgt taatcacggt tgggactcgt tccagatcta ccaaccgctg 4740
gataatagca agtcttacca ggtctacacc aagatgggtc aagcgaagga gaatgatttg 4800
gtgcatggcg atgtggtagt tctggacgga gaacaaatcg ttgctttctt ccgcggcctt 4860
acgctgcgat cagttcctcg tggtgcactg cgtgtcgtcc tgcagactac agtgaaaaag 4920
gccgatcgcc aactaggatt taagacaatg ccgtcgccgc cgcccccgac aacgacaatg 4980
ccaatatcgc cttataaacc agctaatact caggtttcca gccaagctat tccagcagag 5040
gccactcatt ctcacacccc gccacagcca aagcattccc cggtaccgga aactgccgga 5100
agcgctccag cggcaaaagg agtaggtgtc agtaacgaaa agttagatgc tgtaatgcga 5160
gtcgtttcgg aggagagtgg aattgccctc gaggagctca ccgatgacag caactttgct 5220
gacatgggca tcgactctct gagttcaatg gtcattggga gccgcttcag agaggacctg 5280
gggctggacc tggggcctga gttttctctt ttcattgact gcactaccgt gcgtgccttg 5340
aaagacttca tgttgggaag cggggatgct ggcagtggct ccaatgtaga agatcctccc 5400
ccatcagcta ctcccggcat caaccccgaa accgattggt ctagcagtgc ctctgatagt 5460
attttcgcca gcgaagacca cggtcattcg agtgagtccg gcgccgacac cggaagtccg 5520
cctgcacttg atctgaagcc ctactgccgc ccctcaactt ctgtcgtcct acaaggtcta 5580
cctatggtgg cgcggaaaac tctgtttatg ctccctgatg gcgggggggtc tgcgttctcc 5640
tacgcctccc tgccgcgcct caaatcagat actgccgttg tgggcctgaa ttgcccttat 5700
gctcgggatc ccgagaacat gaactgcaca catggagcta tgattgagag cttttgcaat 5760
gagatccggc ggcgacagcc acggggcccc tatcacctgg gcggctggtc gtccggtggt 5820
gcattcgctt acgtcgtggc cgaggcactt gttaaccaag gcgaggaggt gcattcgtta 5880
atcatcattg atgcgcctat tccccaagcc atggaacaac ttccccgagc attttacgag 5940
cactgcaata gcattggatt gttcgctacc cagccggggg ctagtccgga cggctcgact 6000
gagcctccat cctacttaat cccacacttt accgctgtgg tggatgtgat gctggattac 6060
aagctggccc cgttgcatgc gcgccggatg cccaaggtcg gcatcgtctg ggcggcagat 6120
acagtcatgg acgagcggga cgctcccaag atgaaaggaa tgcattttat gattcagaag 6180
cggacggaat ttggtcccga tgggtgggat acgatcatgc ccggggcctc gtttgacatt 6240
gtccgagcag acggtgctaa tcattttacg ttgatgcaaa aggaacatgt ctctataatt 6300
agcgatctga tcgaccgggt catggcttag 6330

<210> 12
<211> 2109
<212> PRT
<213> Aspergillus parasiticus

<400> 12

Met Ala Gln Ser Arg Gln Leu Phe Leu Phe Gly Asp Gln Thr Ala Asp
1 5 10 15

Phe Val Pro Lys Leu Arg Ser Leu Leu Ser Val Gln Asp Ser Pro Ile
20 25 30

Leu Ala Ala Phe Leu Asp Gln Ser His Tyr Val Val Arg Ala Gln Met
35 40 45

Leu Gln Ser Met Asn Thr Val Asp His Lys Leu Ala Arg Thr Ala Asp
50 55 60

Leu Arg Gln Met Val Gln Lys Tyr Val Asp Gly Lys Leu Thr Pro Ala
65 70 75 80

Phe Arg Thr Ala Leu Val Cys Leu Cys Gln Leu Gly Cys Phe Ile Arg
85 90 95

Glu Tyr Glu Glu Ser Gly Asn Met Tyr Pro Gln Pro Ser Asp Ser Tyr
100 105 110

Val Leu Gly Phe Cys Met Gly Ser Leu Ala Ala Val Ala Val Ser Cys
115 120 125

Ser Arg Ser Leu Ser Glu Leu Leu Pro Ile Ala Val Gln Thr Val Leu
130 135 140

Ile Ala Phe Arg Leu Gly Leu Cys Ala Leu Glu Met Arg Asp Arg Val
145 150 155 160

Asp Gly Cys Ser Asp Asp Arg Gly Asp Pro Trp Ser Thr Ile Val Trp
165 170 175

Gly Leu Asp Pro Gln Gln Ala Arg Asp Gln Ile Glu Val Phe Cys Arg
180 185 190

Thr Thr Asn Val Pro Gln Thr Arg Arg Pro Trp Ile Ser Cys Ile Ser
195 200 205

Lys Asn Ala Ile Thr Leu Ser Gly Ser Pro Ser Thr Leu Arg Ala Phe
210 215 220

Cys Ala Met Pro Gln Met Ala Gln His Arg Thr Ala Pro Ile Pro Ile
225 230 235 240

Cys Leu Pro Ala His Asn Gly Ala Leu Phe Thr Gln Ala Asp Ile Thr
245 250 255

Thr Ile Leu Asp Thr Thr Pro Thr Thr Pro Trp Glu Gln Leu Pro Gly
260 265 270

Gln Ile Pro Tyr Ile Ser His Val Thr Gly Asn Val Val Gln Thr Ser
275 280 285

Asn Tyr Arg Asp Leu Ile Glu Val Ala Leu Ser Glu Thr Leu Leu Glu
290 295 300

Gln Val Arg Leu Asp Leu Val Glu Thr Gly Leu Pro Arg Leu Leu Gln
305 310 315 320

Ser Arg Gln Val Lys Ser Val Thr Ile Val Pro Phe Leu Thr Arg Met
325 330 335

Asn Glu Thr Met Ser Asn Ile Leu Pro Asp Ser Phe Ile Ser Thr Glu
340 345 350

Thr Arg Thr Asp Thr Gly Arg Ala Ile Pro Ala Ser Gly Arg Pro Gly
355 360 365

Ala Gly Lys Cys Lys Leu Ala Ile Val Ser Met Ser Gly Arg Phe Pro
370 375 380

Glu Ser Pro Thr Thr Glu Ser Phe Trp Asp Leu Leu Tyr Lys Gly Leu
385 390 395 400

Asp Val Cys Lys Glu Val Pro Arg Arg Arg Trp Asp Ile Asn Thr His
405 410 415

Val Asp Pro Ser Gly Lys Ala Arg Asn Lys Gly Ala Thr Lys Trp Gly
420 425 430

Cys Trp Leu Asp Phe Ser Gly Asp Phe Asp Pro Arg Phe Phe Gly Ile
435 440 445

Ser Pro Lys Glu Ala Pro Gln Met Asp Pro Ala Gln Arg Met Ala Leu
450 455 460

Met Ser Thr Tyr Glu Ala Met Glu Arg Ala Gly Leu Val Pro Asp Thr
465 470 475 480

Thr Pro Ser Thr Gln Arg Asp Arg Ile Gly Val Phe His Gly Val Thr
485 490 495

Ser Asn Asp Trp Met Glu Thr Asn Thr Ala Gln Asn Ile Asp Thr Tyr
500 505 510

Phe Ile Thr Gly Gly Asn Arg Gly Phe Ile Pro Gly Arg Ile Asn Phe
515 520 525

Cys Phe Glu Phe Ala Gly Pro Ser Tyr Thr Asn Asp Thr Ala Cys Ser
530 535 540

Ser Ser Leu Ala Ala Ile His Leu Ala Cys Asn Ser Leu Trp Arg Gly
545 550 555 560

Asp Cys Asp Thr Ala Val Ala Gly Gly Thr Asn Met Ile Tyr Thr Pro
565 570 575

Asp Gly His Thr Gly Leu Asp Lys Gly Phe Phe Leu Ser Arg Thr Gly
580 585 590

Asn Cys Lys Pro Tyr Asp Asp Lys Ala Asp Gly Tyr Cys Arg Ala Glu
595 600 605

Gly Val Gly Thr Val Phe Ile Lys Arg Leu Glu Asp Ala Leu Ala Asp
610 615 620

Asn Asp Pro Ile Leu Gly Val Ile Leu Asp Ala Lys Thr Asn His Ser
625 630 635 640

Ala Met Ser Glu Ser Met Thr Arg Pro His Val Gly Ala Gln Ile Asp
645 650 655

Asn Met Thr Ala Ala Leu Asn Thr Thr Gly Leu His Pro Asn Asp Phe
660 665 670

Ser Tyr Ile Glu Met His Gly Thr Gly Thr Gln Val Gly Asp Ala Val
675 680 685

Glu Met Glu Ser Val Leu Ser Val Phe Ala Pro Ser Glu Thr Ala Arg
690 695 700

Lys Ala Asp Gln Pro Leu Phe Val Gly Ser Ala Lys Ala Asn Val Gly
705 710 715 720

His Gly Glu Gly Val Ser Gly Val Thr Ser Leu Ile Lys Val Leu Met
725 730 735

Met Met Gln His Asp Thr Ile Pro Pro His Cys Gly Ile Lys Pro Gly
740 745 750

Ser Lys Ile Asn Arg Asn Phe Pro Asp Leu Gly Ala Arg Asn Val His
755 760 765

Ile Ala Phe Glu Pro Lys Pro Trp Pro Arg Thr His Thr Pro Arg Arg
770 775 780

Val Leu Ile Asn Asn Phe Ser Ala Ala Gly Gly Asn Thr Ala Leu Ile
785 790 795 800

Val Glu Asp Ala Pro Glu Arg His Trp Pro Thr Glu Lys Asp Pro Arg
805 810 815

Ser Ser His Ile Val Ala Leu Ser Ala His Val Gly Ala Ser Met Lys
820 825 830

Thr Asn Leu Glu Arg Leu His Gln Tyr Leu Leu Lys Asn Pro His Thr
835 840 845

Asp Leu Ala Gln Leu Ser Tyr Thr Thr Thr Ala Arg Arg Trp His Tyr
850 855 860

Leu His Arg Val Ser Val Thr Gly Ala Ser Val Glu Glu Val Thr Arg
865 870 875 880

Lys Leu Glu Met Ala Ile Gln Asn Gly Asp Gly Val Ser Arg Pro Lys
885 890 895

Ser Lys Pro Lys Ile Leu Phe Ala Phe Thr Gly Gln Gly Ser Gln Tyr
900 905 910

Ala Thr Met Gly Lys Gln Val Tyr Asp Ala Tyr Pro Ser Phe Arg Glu
915 920 925

Asp Leu Glu Lys Phe Asp Arg Leu Ala Gln Ser His Gly Phe Pro Ser
930 935 940

Phe Leu His Val Cys Thr Ser Pro Lys Gly Asp Val Glu Glu Met Ala
945 950 955 960

Pro Val Val Val Gln Leu Ala Ile Thr Cys Leu Gln Met Ala Leu Thr
965 970 975

Asn Leu Met Thr Ser Phe Gly Ile Arg Pro Asp Val Thr Val Gly His
980 985 990

Ser Leu Gly Glu Phe Ala Ala Leu Tyr Ala Ala Gly Val Leu Ser Ala
995 1000 1005

Ser Asp Val Val Tyr Leu Val Gly Gln Arg Ala Glu Leu Leu Gln Glu
1010 1015 1020

Arg Cys Gln Arg Gly Thr His Ala Met Leu Ala Val Lys Ala Thr Pro
1025 1030 1035 1040

Glu Ala Leu Ser Gln Trp Ile Gln Asp His Asp Cys Glu Val Ala Cys
1045 1050 1055

Ile Asn Gly Pro Glu Asp Thr Val Leu Ser Gly Thr Thr Lys Asn Val
1060 1065 1070

Ala Glu Val Gln Arg Ala Met Thr Asp Asn Gly Ile Lys Cys Thr Leu
1075 1080 1085

Leu Lys Leu Pro Phe Ala Phe His Ser Ala Gln Val Gln Pro Ile Leu
1090 1095 1100

Asp Asp Phe Glu Ala Leu Ala Gln Gly Ala Thr Phe Ala Lys Pro Gln
1105 1110 1115 1120

Leu Leu Ile Leu Ser Pro Leu Leu Arg Thr Glu Ile His Glu Gln Gly
1125 1130 1135

Val Val Thr Pro Ser Tyr Val Ala Gln His Cys Arg His Thr Val Asp
1140 1145 1150

Met Ala Gln Ala Leu Arg Ser Ala Arg Glu Lys Gly Leu Ile Asp Asp
1155 1160 1165

Lys Thr Leu Val Ile Glu Leu Gly Pro Lys Pro Leu Ile Ser Gly Met
1170 1175 1180

Val Lys Met Thr Leu Gly Asp Lys Ile Ser Thr Leu Pro Thr Leu Ala
1185 1190 1195 1200

Pro Asn Lys Ala Ile Trp Pro Ser Leu Gln Lys Ile Leu Thr Ser Val
1205 1210 1215

Tyr Thr Gly Gly Trp Asp Ile Asn Trp Lys Lys Tyr His Ala Pro Phe
1220 1225 1230

Ala Ser Ser Gln Lys Val Val Asp Leu Pro Ser Tyr Gly Trp Asp Leu
1235 1240 1245

Lys Asp Tyr Tyr Ile Pro Tyr Gln Gly Asp Trp Cys Leu His Arg His
1250 1255 1260

Gln Gln Asp Cys Lys Cys Ala Ala Pro Gly His Glu Ile Lys Thr Ala
1265 1270 1275 1280

Asp Tyr Gln Val Pro Pro Glu Ser Thr Pro His Arg Pro Ser Lys Leu
1285 1290 1295

Asp Pro Ser Lys Glu Ala Phe Pro Glu Ile Lys Thr Thr Thr Thr Leu
1300 1305 1310

His Arg Val Val Glu Glu Thr Thr Lys Pro Leu Gly Ala Thr Leu Val
1315 1320 1325

Val Glu Thr Asp Ile Ser Arg Lys Asp Val Asn Gly Leu Ala Arg Gly
1330 1335 1340

His Leu Val Asp Gly Ile Pro Leu Cys Thr Pro Ser Phe Tyr Ala Asp
1345 1350 1355 1360

Ile Ala Met Gln Val Gly Gln Tyr Ser Met Gln Arg Leu Arg Ala Gly
1365 1370 1375

His Pro Gly Ala Gly Ala Ile Asp Gly Leu Val Asp Val Ser Asp Met
1380 1385 1390

Val Val Asp Lys Ala Leu Val Pro His Gly Lys Gly Pro Gln Leu Leu
1395 1400 1405

Arg Thr Thr Leu Thr Met Glu Trp Pro Pro Lys Ala Ala Ala Thr Thr
1410 1415 1420

Arg Ser Ala Lys Val Lys Phe Ala Thr Tyr Phe Ala Asp Gly Lys Leu
1425 1430 1435 1440

Asp Thr Glu His Ala Ser Cys Thr Val Arg Phe Thr Ser Asp Ala Gln
1445 1450 1455

Leu Lys Ser Leu Arg Arg Ser Val Ser Glu Tyr Lys Thr His Ile Arg
1460 1465 1470

Gln Leu His Asp Gly His Ala Lys Gly Gln Phe Met Arg Tyr Asn Arg
1475 1480 1485

Lys Thr Gly Tyr Lys Leu Met Ser Ser Met Ala Arg Phe Asn Pro Asp
1490 1495 1500

Tyr Met Leu Leu Asp Tyr Leu Val Leu Asn Glu Ala Glu Asn Glu Ala
1505 1510 1515 1520

Ala Ser Gly Val Asp Phe Ser Leu Gly Ser Ser Glu Gly Thr Phe Ala
1525 1530 1535

Ala His Pro Ala His Val Asp Ala Ile Thr Gln Val Ala Gly Phe Ala
1540 1545 1550

Met Asn Ala Asn Asp Asn Val Asp Ile Glu Lys Gln Val Tyr Val Asn
1555 1560 1565

His Gly Trp Asp Ser Phe Gln Ile Tyr Gln Pro Leu Asp Asn Ser Lys
1570 1575 1580

Ser Tyr Gln Val Tyr Thr Lys Met Gly Gln Ala Lys Glu Asn Asp Leu
1585 1590 1595 1600

Val His Gly Asp Val Val Val Leu Asp Gly Glu Gln Ile Val Ala Phe
1605 1610 1615

Phe Arg Gly Leu Thr Leu Arg Ser Val Pro Arg Gly Ala Leu Arg Val
1620 1625 1630

Val Leu Gln Thr Thr Val Lys Lys Ala Asp Arg Gln Leu Gly Phe Lys
1635 1640 1645

Thr Met Pro Ser Pro Pro Pro Pro Thr Thr Thr Met Pro Ile Ser Pro
1650 1655 1660

Tyr Lys Pro Ala Asn Thr Gln Val Ser Ser Gln Ala Ile Pro Ala Glu
1665 1670 1675 1680

Ala Thr His Ser His Thr Pro Pro Gln Pro Lys His Ser Pro Val Pro
1685 1690 1695

Glu Thr Ala Gly Ser Ala Pro Ala Ala Lys Gly Val Gly Val Ser Asn
1700 1705 1710

Glu Lys Leu Asp Ala Val Met Arg Val Val Ser Glu Glu Ser Gly Ile
1715 1720 1725

Ala Leu Glu Glu Leu Thr Asp Asp Ser Asn Phe Ala Asp Met Gly Ile
1730 1735 1740

Asp Ser Leu Ser Ser Met Val Ile Gly Ser Arg Phe Arg Glu Asp Leu
1745 1750 1755 1760

Gly Leu Asp Leu Gly Pro Glu Phe Ser Leu Phe Ile Asp Cys Thr Thr
1765 1770 1775

Val Arg Ala Leu Lys Asp Phe Met Leu Gly Ser Gly Asp Ala Gly Ser
1780 1785 1790

Gly Ser Asn Val Glu Asp Pro Pro Pro Ser Ala Thr Pro Gly Ile Asn
1795 1800 1805

Pro Glu Thr Asp Trp Ser Ser Ser Ala Ser Asp Ser Ile Phe Ala Ser
1810 1815 1820

Glu Asp His Gly His Ser Ser Glu Ser Gly Ala Asp Thr Gly Ser Pro
1825 1830 1835 1840

Pro Ala Leu Asp Leu Lys Pro Tyr Cys Arg Pro Ser Thr Ser Val Val
1845 1850 1855

Leu Gln Gly Leu Pro Met Val Ala Arg Lys Thr Leu Phe Met Leu Pro
1860 1865 1870

Asp Gly Gly Gly Ser Ala Phe Ser Tyr Ala Ser Leu Pro Arg Leu Lys
1875 1880 1885

Ser Asp Thr Ala Val Val Gly Leu Asn Cys Pro Tyr Ala Arg Asp Pro
1890 1895 1900

Glu Asn Met Asn Cys Thr His Gly Ala Met Ile Glu Ser Phe Cys Asn
1905 1910 1915 1920

Glu Ile Arg Arg Arg Gln Pro Arg Gly Pro Tyr His Leu Gly Gly Trp
1925 1930 1935

Ser Ser Gly Gly Ala Phe Ala Tyr Val Val Ala Glu Ala Leu Val Asn
1940 1945 1950

Gln Gly Glu Glu Val His Ser Leu Ile Ile Ile Asp Ala Pro Ile Pro
1955 1960 1965

Gln Ala Met Glu Gln Leu Pro Arg Ala Phe Tyr Glu His Cys Asn Ser
1970 1975 1980

Ile Gly Leu Phe Ala Thr Gln Pro Gly Ala Ser Pro Asp Gly Ser Thr
1985 1990 1995 2000

Glu Pro Pro Ser Tyr Leu Ile Pro His Phe Thr Ala Val Val Asp Val
2005 2010 2015

Met Leu Asp Tyr Lys Leu Ala Pro Leu His Ala Arg Arg Met Pro Lys
2020 2025 2030

Val Gly Ile Val Trp Ala Ala Asp Thr Val Met Asp Glu Arg Asp Ala
2035 2040 2045

Pro Lys Met Lys Gly Met His Phe Met Ile Gln Lys Arg Thr Glu Phe
2050 2055 2060

Gly Pro Asp Gly Trp Asp Thr Ile Met Pro Gly Ala Ser Phe Asp Ile
2065 2070 2075 2080

Val Arg Ala Asp Gly Ala Asn His Phe Thr Leu Met Gln Lys Glu His
2085 2090 2095

Val Ser Ile Ile Ser Asp Leu Ile Asp Arg Val Met Ala
2100 2105

<210> 13
<211> 1986
<212> PRT
<213> Aspergillus nidulans

<400> 13
Met Glu Asp Pro Tyr Arg Val Tyr Leu Phe Gly Asp Gln Thr Gly Asp
1 5 10 15

Phe Glu Val Gly Leu Arg Arg Leu Leu Gln Ala Lys Asn His Ser Leu
20 25 30

Leu Ser Ser Phe Leu Gln Arg Ser Tyr His Ala Val Arg Gln Glu Ile
35 40 45

Ser His Leu Pro Pro Ser Glu Arg Ser Thr Phe Pro Arg Phe Thr Ser
50 55 60

Ile Gly Asp Leu Leu Ala Arg His Cys Glu Ser Pro Gly Asn Pro Ala
65 70 75 80

Ile Glu Ser Val Leu Thr Cys Ile Tyr Gln Leu Gly Cys Phe Ile Asn
85 90 95

Tyr Tyr Gly Asp Leu Gly His Thr Phe Pro Ser His Ser Gln Ser Gln
100 105 110

Leu Val Gly Leu Cys Thr Gly Leu Leu Ser Cys Ala Ala Val Ser Cys
115 120 125

Ala Ser Asn Ile Gly Glu Leu Leu Lys Pro Ala Val Glu Val Val Val
130 135 140

Val Ala Leu Arg Leu Gly Leu Cys Val Tyr Arg Val Arg Lys Leu Phe
145 150 155 160

Gly Gln Asp Gln Ala Ala Pro Leu Ser Trp Ser Ala Leu Val Ser Gly
165 170 175

Leu Ser Glu Ser Glu Gly Thr Ser Leu Ile Asp Lys Phe Thr Arg Arg
180 185 190

Asn Val Ile Pro Pro Ser Ser Arg Pro Tyr Ile Ser Ala Val Cys Ala
195 200 205

Asn Thr Leu Thr Ile Ser Gly Pro Pro Val Val Leu Asn Gln Phe Leu
210 215 220

Asp Thr Phe Ile Ser Gly Lys Asn Lys Ala Val Met Val Pro Ile His
225 230 235 240

Gly Pro Phe His Ala Ser His Leu Tyr Glu Lys Arg Asp Val Glu Trp
245 250 255

Ile Leu Lys Ser Cys Asn Val Glu Thr Ile Arg Asn His Lys Pro Arg
260 265 270

Ile Pro Val Leu Ser Ser Asn Thr Gly Glu Leu Ile Val Val Glu Asn
275 280 285

Met Glu Gly Phe Leu Lys Ile Ala Leu Glu Glu Ile Leu Leu Arg Gln
290 295 300

Met Ser Trp Asp Lys Val Thr Asp Ser Cys Ile Ser Ile Leu Lys Ser
305 310 315 320

Val Gly Asp Asn Lys Pro Lys Lys Leu Leu Pro Ile Ser Ser Thr Ala
325 330 335

Thr Gln Ser Leu Phe Asn Ser Leu Lys Lys Ser Asn Leu Val Asn Ile
340 345 350

Glu Val Asp Gly Gly Ile Ser Asp Phe Ala Ala Glu Thr Gln Leu Val
355 360 365

Asn Gln Thr Gly Arg Ala Glu Leu Ser Lys Ile Ala Ile Ile Gly Met
370 375 380

Ser Gly Arg Phe Pro Glu Ala Asp Ser Pro Gln Asp Phe Trp Asn Leu
385 390 395 400

Leu Tyr Lys Gly Leu Asp Val His Arg Lys Val Pro Glu Asp Arg Trp
405 410 415

Asp Ala Asp Ala His Val Asp Leu Thr Gly Thr Ala Thr Asn Thr Ser
420 425 430

Lys Val Pro Tyr Gly Cys Trp Ile Arg Glu Pro Gly Leu Phe Asp Pro
435 440 445

Arg Phe Phe Asn Met Ser Pro Arg Glu Ala Leu Gln Ala Asp Pro Ala
450 455 460

Gln Arg Leu Ala Leu Leu Thr Ala Tyr Glu Ala Leu Glu Gly Ala Gly
465 470 475 480

Phe Val Pro Asp Ser Thr Pro Ser Thr Gln Arg Asp Arg Val Gly Ile
485 490 495

Phe Tyr Gly Met Thr Ser Asp Asp Tyr Arg Glu Val Asn Ser Gly Gln
500 505 510

Asp Ile Asp Thr Tyr Phe Ile Pro Gly Gly Asn Arg Ala Phe Thr Pro
515 520 525

Gly Arg Ile Asn Tyr Tyr Phe Lys Phe Ser Gly Pro Ser Val Ser Val
530 535 540

Asp Thr Ala Cys Ser Ser Ser Leu Ala Ala Ile His Leu Ala Cys Asn
545 550 555 560

Ser Ile Trp Arg Asn Asp Cys Asp Thr Ala Ile Thr Gly Gly Val Asn
565 570 575

Ile Leu Thr Asn Pro Asp Asn His Ala Gly Leu Asp Arg Gly His Phe
580 585 590

Leu Ser Arg Thr Gly Asn Cys Asn Thr Phe Asp Asp Gly Ala Asp Gly
595 600 605

Tyr Cys Arg Ala Asp Gly Val Gly Thr Val Val Leu Lys Arg Leu Glu
610 615 620

Asp Ala Leu Ala Asp Asn Asp Pro Ile Leu Gly Val Ile Asn Gly Ala
625 630 635 640

Tyr Thr Asn His Ser Ala Glu Ala Val Ser Ile Thr Arg Pro His Val
645 650 655

Gly Ala Gln Ala Phe Ile Phe Lys Lys Leu Leu Asn Glu Ala Asn Val
660 665 670

Asp Pro Lys Asn Ile Ser Tyr Ile Glu Met His Gly Thr Gly Thr Gln
675 680 685

Ala Gly Asp Ala Val Glu Met Gln Ser Val Leu Asp Val Phe Ala Pro
690 695 700

Asp His Arg Arg Gly Pro Gly Gln Ser Leu His Leu Gly Ser Ala Lys
705 710 715 720

Ser Asn Ile Gly His Gly Glu Ser Ala Ser Gly Val Thr Ser Leu Val
725 730 735

Lys Val Leu Leu Met Met Lys Glu Asn Met Ile Pro Pro His Cys Gly
740 745 750

Ile Lys Thr Lys Ile Asn His Asn Phe Pro Thr Asp Leu Ala Gln Arg
755 760 765

Asn Val His Ile Ala Leu Gln Pro Thr Ala Trp Asn Arg Pro Ser Phe
770 775 780

Gly Lys Arg Gln Ile Phe Leu Asn Asn Phe Ser Ala Ala Gly Gly Asn
785 790 795 800

Thr Ala Leu Leu Leu Glu Asp Gly Pro Val Ser Asp Pro Glu Gly Glu
805 810 815

Asp Lys Arg Arg Thr His Val Ile Thr Leu Ser Ala Arg Ser Gln Thr
820 825 830

Ala Leu Gln Asn Asn Ile Asp Ala Leu Cys Gln Tyr Ile Ser Glu Gln
835 840 845

Glu Lys Thr Phe Gly Val Lys Asp Ser Asn Ala Leu Pro Ser Leu Ala
850 855 860

Tyr Thr Thr Thr Ala Arg Arg Ile His His Pro Phe Arg Val Thr Ala
865 870 875 880

Ile Gly Ser Ser Phe Gln Glu Met Arg Asp Ser Leu Ile Ala Ser Ser
885 890 895

Arg Lys Glu Phe Val Ala Val Pro Ala Lys Thr Pro Gly Ile Gly Phe
900 905 910

Leu Phe Thr Gly Gln Gly Ala Gln Tyr Ala Ala Met Gly Lys Gln Leu
915 920 925

Tyr Glu Asp Cys Ser His Phe Arg Ser Ala Ile Glu His Leu Asp Cys
930 935 940

Ile Ser Gln Gly Gln Asp Leu Pro Ser Ile Leu Pro Leu Val Asp Gly
945 950 955 960

Ser Leu Pro Leu Ser Glu Leu Ser Pro Val Val Val Gln Leu Gly Thr
965 970 975

Thr Cys Val Gln Met Ala Leu Ser Ser Phe Trp Ala Ser Leu Gly Ile
980 985 990

Thr Pro Ser Phe Val Leu Gly His Ser Leu Gly Asp Phe Ala Ala Met
995 1000 1005

Asn Ala Ala Gly Val Leu Ser Thr Ser Asp Thr Ile Tyr Ala Cys Gly
1010 1015 1020

Arg Arg Ala Gln Leu Leu Thr Glu Arg Cys Gln Pro Gly Thr His Ala
1025 1030 1035 1040

Met Leu Ala Ile Lys Ala Pro Leu Val Glu Val Lys Gln Leu Leu Asn
1045 1050 1055

Glu Lys Val His Asp Met Ala Cys Ile Asn Ser Pro Ser Glu Thr Val
1060 1065 1070

Ile Ser Gly Pro Lys Ser Ser Ile Asp Glu Leu Ser Arg Ala Cys Ser
1075 1080 1085

Glu Lys Gly Leu Lys Ser Thr Ile Leu Thr Val Pro Tyr Ala Phe His
1090 1095 1100

Ser Ala Gln Val Glu Pro Ile Leu Glu Asp Leu Glu Lys Ala Leu Gln
1105 1110 1115 1120

Gly Ile Thr Phe Asn Lys Pro Ser Val Pro Phe Val Ser Ala Leu Leu
1125 1130 1135

Gly Glu Val Ile Thr Glu Ala Gly Ser Asn Ile Leu Asn Ala Glu Tyr
1140 1145 1150

Leu Val Arg His Cys Arg Glu Thr Val Asn Phe Leu Ser Ala Phe Glu
1155 1160 1165

Ala Val Arg Asn Ala Lys Leu Gly Gly Asp Gln Thr Leu Trp Leu Glu
1170 1175 1180

Val Gly Pro His Thr Val Cys Ser Gly Met Val Lys Ala Thr Leu Gly
1185 1190 1195 1200

Pro Gln Thr Thr Thr Met Ala Ser Leu Arg Arg Asp Glu Asp Thr Trp
1205 1210 1215

Lys Val Leu Ser Asn Ser Leu Ser Ser Leu Tyr Leu Ala Gly Val Asp
1220 1225 1230

Ile Asn Trp Lys Gln Tyr His Gln Asp Phe Ser Ser Ser His Arg Val
1235 1240 1245

Leu Pro Leu Pro Thr Tyr Lys Trp Asp Leu Lys Asn Tyr Trp Ile Pro
1250 1255 1260

Tyr Arg Asn Asn Phe Cys Leu Thr Lys Gly Ser Ser Met Ser Ala Ala
1265 1270 1275 1280

Ser Ala Ser Leu Gln Pro Thr Phe Leu Thr Thr Ser Ala Gln Arg Val
1285 1290 1295

Val Glu Ser Arg Asp Asp Gly Leu Thr Ala Thr Val Val Val His Asn
1300 1305 1310

Asp Ile Ala Asp Pro Asp Leu Asn Arg Val Ile Gln Gly His Lys Val
1315 1320 1325

Asn Gly Ala Ala Leu Cys Pro Ser Ser Leu Tyr Ala Asp Ser Ala Gln
1330 1335 1340

Thr Leu Ala Glu Tyr Leu Ile Glu Lys Tyr Lys Pro Glu Leu Lys Gly
1345 1350 1355 1360

Ser Gly Leu Asp Val Cys Asn Val Thr Val Pro Lys Pro Leu Ile Ala
1365 1370 1375

Lys Thr Gly Lys Glu Gln Phe Arg Ile Ser Ala Thr Ala Asn Trp Val
1380 1385 1390

Asp Lys His Val Ser Val Gln Val Phe Ser Val Thr Ala Glu Gly Lys
1395 1400 1405

Lys Leu Ile Asp His Ala His Cys Glu Val Lys Leu Phe Asp Cys Met
1410 1415 1420

Ala Ala Asp Leu Glu Trp Lys Arg Gly Ser Tyr Leu Val Lys Arg Ser
1425 1430 1435 1440

Ile Glu Leu Leu Glu Asn Ser Ala Val Lys Gly Asp Ala His Arg Leu
1445 1450 1455

Arg Arg Gly Met Val Tyr Lys Leu Phe Ser Ala Leu Val Asp Tyr Asp
1460 1465 1470

Glu Asn Tyr Gln Ser Ile Arg Glu Val Ile Leu Asp Ser Glu His His
1475 1480 1485

Glu Ala Thr Ala Leu Val Lys Phe Gln Ala Pro Gln Ala Asn Phe His
1490 1495 1500

Arg Asn Pro Tyr Trp Ile Asp Ser Phe Gly His Leu Ser Gly Phe Ile
1505 1510 1515 1520

Met Asn Ala Ser Asp Gly Thr Asp Ser Lys Ser Gln Val Phe Val Asn
1525 1530 1535

His Gly Trp Asp Ser Met Arg Cys Leu Lys Lys Phe Ser Ala Asp Val
1540 1545 1550

Thr Tyr Arg Thr Tyr Val Arg Met Gln Pro Trp Arg Asp Ser Ile Trp
1555 1560 1565

Ala Gly Asn Val Tyr Ile Phe Glu Gly Asp Asp Ile Ile Ala Val Phe
1570 1575 1580

Gly Gly Val Lys Phe Gln Ala Leu Ser Arg Lys Ile Leu Asp Ile Ala
1585 1590 1595 1600

Leu Pro Pro Ala Gly Leu Ser Lys Ala Gln Thr Ser Pro Ile Gln Ser
1605 1610 1615

Ser Ala Pro Gln Lys Pro Ile Glu Thr Ala Lys Pro Thr Ser Arg Pro
1620 1625 1630

Ala Pro Pro Val Thr Met Lys Ser Phe Val Lys Lys Ser Ala Gly Pro
1635 1640 1645

Ser Val Val Val Arg Ala Leu Asn Ile Leu Ala Ser Glu Val Gly Leu
1650 1655 1660

Ser Glu Ser Asp Met Ser Asp Asp Leu Val Phe Ala Asp Tyr Gly Val
1665 1670 1675 1680

Asp Ser Leu Leu Ser Leu Thr Val Thr Gly Lys Tyr Arg Glu Glu Leu
1685 1690 1695

Asn Leu Asp Met Asp Ser Ser Val Phe Ile Glu His Pro Thr Val Gly
1700 1705 1710

Asp Phe Lys Arg Phe Val Thr Gln Leu Ser Pro Ser Val Ala Ser Asp
1715 1720 1725

Ser Ser Ser Thr Asp Arg Glu Ser Glu Tyr Ser Phe Asn Gly Asp Ser
1730 1735 1740

Cys Ser Gly Leu Ser Ser Pro Ala Ser Pro Gly Thr Val Ser Pro Pro
1745 1750 1755 1760

Asn Glu Lys Val Ile Gln Ile His Glu Asn Gly Thr Met Lys Glu Ile
1765 1770 1775

Arg Ala Ile Ile Ala Asp Glu Ile Gly Val Ser Ala Asp Glu Ile Lys
1780 1785 1790

Ser Asp Glu Asn Leu Asn Glu Leu Gly Met Asp Ser Leu Leu Ser Leu
1795 1800 1805

Thr Val Leu Gly Lys Ile Arg Glu Ser Leu Asp Met Asp Leu Pro Gly
1810 1815 1820

Glu Phe Phe Ile Glu Asn Gln Thr Leu Asp Gln Ile Glu Thr Ala Leu
1825 1830 1835 1840

Asp Leu Lys Pro Lys Ala Val Pro Thr Ala Val Pro Gln Ser Gln Pro
1845 1850 1855

Ile Thr Leu Pro Gln Ser Gln Ser Thr Lys Gln Leu Ser Thr Arg Pro
1860 1865 1870

Thr Ser Ser Ser Asp Asn His Pro Pro Ala Thr Ser Ile Leu Leu Gln
1875 1880 1885

Gly Asn Pro Arg Thr Ala Ser Lys Thr Leu Phe Leu Phe Pro Asp Gly
1890 1895 1900

Ser Gly Ser Ala Thr Ser Tyr Ala Thr Ile Pro Gly Val Ser Pro Asn
1905 1910 1915 1920

Val Ala Val Tyr Gly Leu Asn Cys Pro Tyr Met Lys Ala Pro Glu Lys
1925 1930 1935

Leu Thr Cys Ser Leu Asp Ser Leu Thr Thr Pro Tyr Leu Ala Glu Ile
1940 1945 1950

Arg Arg Arg Gln Pro Thr Gly Pro Tyr Asn Leu Gly Gly Trp Ser Gln
1955 1960 1965

Ala Gly Ser Ala His Thr Thr Arg His Ala Ser Ser Tyr Cys Ser Arg
1970 1975 1980

Ala Lys
1985

<210> 14
<211> 53
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 14
atgaagcttg gggtttgagg gccaatggaa cgaaactagt gtaccacttg acc 53

<210> 15
<211> 28
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 15
gacagatctg gcgccattcg ccattcag 28

<210> 16
<211> 20
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 16
ggaatcggtc aatacactac 20

<210> 17
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 17
tgtagatctc tattcctttg ccctcggacg agt 33

<210> 18
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 18
ggccgccacg gatatcttgg ccaaagaatt cctgg 35

<210> 19
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 19
cggtgcctat agaaccggtt tcttaaggac cgcgc 35

<210> 20
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 20
gayccmgtty ttyaayatg 19

<210> 21
<211> 17
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 21
gtccgtccrt gcatytc 17

<210> 22
<211> 34
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 22
ataagaatgc ggccgcaatg gccctcgaaa cagc 34

<210> 23
<211> 29
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 23
aaatggcgcg ccgcgcccag aatgacacc 29

<210> 24
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 24
tgccacctgt agtctgcaat cag 23

<210> 25
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 25
tgactaaccc tgacaacttc gctg 24

<210> 26
<211> 19
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 26
ccaggatccg actgctcag 19

<210> 27
<211> 21
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 27
ctacatcgag atgcacggca c 21

<210> 28
<211> 16
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 28
ngtcgaswga nawgaa 16

<210> 29
<211> 16
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 29
gtncgaswca nawgtt 16

<210> 30
<211> 16
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 30
wgtgnagwan canaga 16

<210> 31
<211> 15
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 31
ntcgastwts gwgtt 15

<210> 32
<211> 16
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 32
tgwgnagwan casaga 16

<210> 33
<211> 16
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 33
agwgnagwan cawagg 16

<210> 34
<211> 14
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 34
cawcgcngaa sgaa 14

<210> 35
<211> 14
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 35
tcstcgnact wgga 14

<210> 36
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 36
ttgttactgg agaggtaatg aag 23

<210> 37
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 37
tgagacagat ctcgcgagcc ctc 23

<210> 38
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 38
atgtctccaa aggaagctga gc 22

<210> 39
<211> 22
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 39
tcgagtgatg gatactgctt cg 22

<210> 40
<211> 29
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 40
cggctacact agaaggacag tatttggta 29

<210> 41
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 41
gtcaggcaac tatggatgaa cgaaatagac 30

<210> 42
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 42
acccatctca taaataacgt catgc 25

<210> 43
<211> 23
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 43
caactctatc agagcttggt tga 23

<210> 44
<211> 30
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 44
cccgaattca tgagctttgt tcaaataagg 30

<210> 45
<211> 39
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 45
ttattctaga ttttccatgg gaatggatac agtcttacg 39

<210> 46
<211> 33
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 46
cgccaccatg gtgagcaagg gcgaggagct gtt 33

<210> 47
<211> 39
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 47
tatgatctag agtcgcggcc gctttacttg tacagctcg 39

<210> 48
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 48
gcttctaatc cgtactagtg gatca 25

<210> 49
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer

<400> 49
ctttgatctt ttctacgggg tctga 25